Behavioural reactions of cod (Gadus morhua) and plaice (Pleuronectes platessa) to sound resembling offshore wind turbine noise

Dissertation

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Abbreviations and Acronyms used in the Thesis

BEOFINO	Ökologische Begleitforschung zur Windenergienutzung im Offshore-Bereich auf Forschungsplattformen in der Nord- und Ostsee (Ecological research on the impact of Offshore Wind farms based on research platforms in the North and Baltic Sea)
BfN	Bundesamt für Naturschutz (Federal Agency for Nature Conservation)
B_{lim}	limit biomass
B_{pa}	precautionary biomass
cacodylate buffer	sodium cacodylate trihydrate buffer
dB	decibel
dB re 1 µPa	decibel relative to a reference of 1 micro Pascal
e.g.	for example (Latin exempli gratia)
et al.	and others (Latin et alii)
FRS Marlab	Fisheries Research Services Marine Laboratory
GW	Gigawatt (1,000,000,000 Watt)
h	hour
H	hydrophone
Hz	Hertz
IfaF	Institut für angewandte Fischbiologie (Institute for applied fish biology)
IfaÖ	Institut für angewandte Ökologie (Institute for applied Ecology)
ITC	International Transducer Corporation
KHz	Kilohertz
kW	Kilowatt
lg	logarithm
LS	loudspeaker
MINOS	Forschungsprojekt Marine Warmblüter in Nord- und Ostsee (Research project Marine warm-blooded animals in the North and Baltic Seas)
MW	Megawatt (1,000,000 Watt)
pers.	personal
rms	root mean square
S.D.	standard deviation
SSB	spawning stock biomass
TL	total length
UK	United Kingdom
USRL	Underwater Sound Reference Laboratory, US Navy
UV	ultraviolet

Summary

Effects of low frequency sound such as emitted by offshore wind turbines on fish were investigated in an annular tank with 10 m diameter filled with sea water 1.26 m deep. The tank was divided in neighbouring quarters by sound barriers, so that sound pressure differences of 32 to 52 dB were achieved. The experimental fish were free to move around in the tank and therefore could avoid high sound levels. Spontaneously, the fish preferred one of the quarters. Therefore the experimental sound was produced in that quarter by an underwater loudspeaker to test whether the fish would leave that quarter during sound production.

For the investigations, cod and plaice were chosen as two important North and Baltic Sea species with differently pronounced hearing abilities. In both species, juveniles and adults were tested. With every group of fish, 9 to 10 experiments were done, consisting of 24 hours continuous pure tone production followed by 5 days of recovery. Five frequencies between 25 and 250 Hz and sound levels of 130 and 140 dB re 1 µPa were chosen.

The 24 hour periods before, during, and after sound production were evaluated by registering the fish number, their distribution and their behaviour in the tank quarter containing the sound source, using continuous video surveillance with overhead cameras.

Cod

15 juvenile (32 - 53 cm TL) and 13 adult (56 - 72 cm TL) cod were tested. Activity was higher in daytime, which was more pronounced in juvenile cod. Except for 250 Hz, in most experiments significantly less fish were observed in the preferred quarter during sound production, than in the periods before and after, but the quarter was not left completely. Reactions were most pronounced at 60 Hz and 90 Hz.

From these results escape in some measure of cod from sound in the vicinity of offshore wind farms would be expected. The one day duration of the sound does not allow conclusions, however, whether the escape would be permanent.

Plaice

20 juvenile (24 - 32 cm TL) and 20 adult (26 - 43 cm TL) plaice were tested. Without sound, the preference for one tank quarter was very strong in adults and less pronounced in juveniles. The fish showed a diel rhythm with higher activity in daytime, which was more obvious in adults than in juveniles. During sound production reactions were inconsistent in that both avoidance or attraction were found. Dependence on frequency or sound level was not found, nor was complete avoidance of the preferred quarter.

When specimens were added to the tank while the sound was being played in the preferred quarter settlement was delayed.

The results indicate that plaice detected the sound but the type of reaction was variable. They suggest that permanent avoidance of offshore wind farm areas would not be expected.

Reaction thresholds

Behavioural reactions could be observed at sound levels of less than 30 dB above the detection threshold. The results are discussed in connection with different thresholds presented by other authors indicating the urgent need for further research to define reliable reaction thresholds.

Deutsche Zusammenfassung

Die Auswirkungen von tieffrequentem, den Emissionen von Windfarmen ähnelndem Schall auf Fische wurden in einem ringförmigen Versuchsbecken mit 10 m Durchmesser und 1,26 m Tiefe untersucht. Das Versuchbecken war mit schallisolierenden Wänden in verbundene Viertel unterteilt, was zu Schalldruckdifferenzen von 32 bis 52 dB im Becken führte. Die Versuchsfische konnten sich frei im Becken bewegen und so hohen Schalldruckpegeln ausweichen. Aufgrund einer Präferenz der Fische für ein bestimmtes Beckenviertel erfolgte die Schallproduktion in diesem Viertel.

Die Experimente wurden mit Scholle und Kabeljau als zwei wichtigen Nord- und Ostseearten mit unterschiedlich ausgeprägtem Hörvermögen in zwei verschiedenen Altersklassen (juvenil und adult) durchgeführt. Jede Versuchsfischgruppe durchlief eine Reihe von 9 bis 10 Einzelversuchen, die jeweils aus einer 24-stündigen Beschallung und fünf Tagen Erholungsphase bestanden. Schall wurde in fünf Frequenzen zwischen 25 und 250 Hz mit Schalldruckpegeln von 130 und 140 dB re 1µPa produziert.

Mit Hilfe von Videokameras über dem Versuchsbecken wurden die Fische im Becken überwacht. Ausgewertet wurde die Anzahl, Verteilung und das Verhalten der Fische im bevorzugten Beckenviertel in den 24 Stunden vor, während und nach Schallproduktion.

Kabeljau

Es wurden 15 juvenile (32 - 53 cm TL) und 13 adulte (56 - 72 cm TL) Kabeljau getestet. Die Aktivität war am Tag höher, was bei juvenilen Kabeljau stärker ausgeprägt war. Außer bei der Frequenz 250 Hz zeigten sich in vielen der Versuche deutliche Reaktionen auf den Schall mit signifikant weniger Fischen im bevorzugten Beckenviertel im Vergleich zu den Phasen ohne Beschallung. Jedoch verließen nicht alle Tiere das beschallte Beckenviertel. Am stärksten waren die Reaktionen in den Versuchen mit 60 und 90 Hz.

Die Ergebnisse lassen schallbedingte Vermeidung von Gebieten in und um Windparks durch Kabeljau erwarten. Eine Einschätzung über mögliche Gewöhnung an den Schall kann aus den vorliegenden Ergebnissen nicht abgeleitet werden.

Scholle

Es wurden 20 juvenile (24 - 32 cm TL) und 20 adulte (26 - 43 cm TL) Schollen getestet. Ohne Beschallung zeigten besonders die adulten aber auch die juvenilen Schollen eine stark ausgeprägte Präferenz für ein Beckenviertel. Schollen zeigten einen Tag-Nacht-Rhythmus mit höherer Aktivität am Tag, der bei den Adulten stärker ausgeprägt war als bei den Juvenilen. Auf den Schall reagierten die Schollen uneinheitlich mit sowohl Vermeidung als auch Attraktion. Es zeigte sich keine klare Abhängigkeit des Verhaltens, weder von Frequenzen noch von Schalldruckpegeln, und auch keine vollständige Meidung des bevorzugten Beckenviertels.

Wurden Schollen während Schallproduktion ins Versuchsbecken eingesetzt, zeigte sich eine verzögerte Ansiedlung im bevorzugten Beckenviertel.

Die Ergebnisse deuten darauf hin, dass Schollen den Schall wahrnehmen können, ihn aber nicht durchgängig meiden.

Reaktionsgrenzen

Es konnten Verhaltensänderungen auf Schall, der weniger als 30 dB oberhalb der Wahrnehmungsgrenze lag, nachgewiesen werden. Im Vergleich mit den von anderen Autoren angegebenen Reaktionsgrenzen ergibt sich dringender Forschungsbedarf um verlässliche Grenzwerte definieren zu können.

1 Introduction

1.1 Project overview

While the increased use of offshore wind energy is desirable for climate protection, offshore wind farms will have an impact on the marine life in the vicinity of the turbines as artificial structures may attract different species and will influence sediment movements, noise will be emitted and electro magnetic field changes will appear in the direct vicinity of the cables (ELSAM ENGINEERING & ENERGI E2 A/S 2005). For this reason studies on the influence of offshore wind farms on marine life are necessary to identify potential disturbance of marine organism and birds in order to minimize these effects. A number of studies have investigated the influence of offshore wind farms on the marine environment such as MINOS (marine mammals and birds) and BEOFINO (benthos) funded by the German Government (MÜNTER 2004).

Awareness of the vulnerability of marine mammals to man-made noise is relatively high in science, public, industry and government agencies and is the subject of a number of research projects and publications (e.g. RICHARDSON et al. 1995, ERBE & FARMER 2000, SOUTHALL ET AL. 2000, TEILMANN ET AL. 2002, HAMMOND ET AL. 2002, KOSCHINSKI ET AL. 2003).

Interest in the influence of anthropogenic sound on fish has mainly been restricted to the scientific community and hardly discussed in public. A number of studies have been carried out, some of which have included wind farm noise specifically (see section 1.2.4.4).

Research on potential disturbance of fish is important for fisheries that already suffer from low catch rates caused by decreasing fish stocks in Atlantic, North and Baltic Sea (ZIMMERMANN & GRÖHSLER 2004). But especially in fish populations it is difficult to examine disturbing factors in the open sea. Reactions in the wild are a combined effect of a number of variable factors and the size of fish stocks is varying largely with time (ICES 2006, HOFFMANN et al. 2000, KARASIOVA & ZEZERA 2005).

One subject of concern is low frequency sound emitted by the offshore turbine pile during operation. Noise emitted by offshore wind farms could stress animals or might disturb communication. Sound is an important means of orientation and communication for many marine animals especially in low light levels and impaired vision (TAVOLGA 1974, KRAAN & VAN ETTEN 1995). Sound can be transferred fast and far underwater (HAWKINS & MYRBERG JR 1983) and low frequencies can travel over extremely long distances (WILLE 1986).

In the open sea it is difficult to examine whether sound emissions have an impact on the behaviour of fish for a number of reasons:

- the sound cannot be isolated from other biotic and abiotic factors influencing behaviour
- Offshore wind farms with turbine sizes like those planned in the North and Baltic Seas are not yet built.
- Offshore field experiments are logistically difficult and expensive to carry out

Therefore, fish were tested in tank-based experiments where a controlled sound pressure difference in defined sound fields could be created. As model species plaice and cod were chosen as representatives for North and Baltic Seas species with differently pronounced hearing abilities.

1.2 General background and state of research

1.2.1 Offshore wind technology in Europe

The number of offshore wind farms planned or already built in European waters has been increasing rapidly over the last ten years (SUNBEAM 2006). At the end of 2004 offshore wind farms totalling approximately 700 MW were installed in Europe, mainly in Danish and UK waters (SUNBEAM 2006). At the same time more than 43,000 MW offshore wind farm capacity was planned for European waters. Existing large scale wind farms as Horns Rev (Denmark, 80 turbines) and North Hoyle (UK, 30 turbines) contain turbines of 2 MW. A few larger turbines of up to 4.5 MW have been tested but are not ready for large scale wind farms yet. Information about the mentioned wind farms are given in Table A 1 (appendix).

The capacity of wind energy worldwide reached a total of more than 58,500 MW by the end of 2005, of which nearly 70% was from Europe (DEWI 2006). Worldwide, wind energy currently delivers 1% of electricity. It is expected that an overall capacity of 132,000 MW will be in place by 2010 (DEWI 2006) and the European Wind Energy Association has set a target of 70 GW wind energy installed offshore by 2020 (GREENPEACE 2005). Wind farms need to be located in areas with suitably high amounts of wind. Their construction can be contentious due to issues of noise emission and impact of the turbines on the landscape. Being densely populated, the capacity for onshore wind farms in Europe is restricted. At sea, wind speeds are considerably higher and more predictable than onshore. At most of the suitable offshore sites in northern European waters, the wind is expected to deliver between 20% and 40% more energy than on good onshore sites (GREENPEACE 2005). On the other hand the construction of offshore wind farms and the connection to the onshore cable grid is more expensive.

1.2.2 Acoustic field in offshore wind farms

Not many sound measurements have been carried out at offshore wind turbines and those that exist are mostly confined to single turbines (e.g. WESTERBERG 2000, DEGN 2000, ENGELL-SØRENSEN 2002). The sound spectrum close to a sound source differs from the spectra at greater distances since different frequencies are emitted at different sound levels. Therefore, not only the wavelength but also the sound level of single frequencies alters the frequency spectrum of the sound with the distance. The sound field produced by a single turbine will differ from the sound field produced by multiple turbines. With distances of some hundred meters (e.g. Robin Rigg/Scotland, 450 m, Nysted (Rødsand)/ Denmark, 850 m) to each other and a large number of turbines, wind farms become a large-scale sound source influencing not only the wind farm area but also the surroundings. Sound waves of single turbines can join together and interferences can occur with local sound pressure peaks and dips. Sound levels and frequency spectra of offshore wind turbine emissions will depend on the turbine type, the foundation, water depth and seabed conditions. DEGN (2000) predicted sound emissions for 2-MW turbines with concrete or monopile foundations (Fig. A 1 and Fig. A 2, appendix) and expected higher sound levels in frequencies below 50 Hz using concrete compared with the monopile foundations. Measurements of sound emissions of offshore wind turbines exist for smaller turbines such as the 1.5-MW turbines in Utgrunden (Sweden) (INGEMANSSON 2003, Fig. A 3, appendix). The sound spectrum of a 4.5-MW offshore turbine with monopile foundation has been predicted (DEWI 2004) to give sound levels with a maximum of about 140 dB re 1µPa at 130 Hz and 200 Hz.

Apart from turbine and location characteristics, the sound varies with the wind speed causing increasing sound levels and higher emission in higher frequencies due to increased turbine rotational speed (INGEMANSSON 2003). At the same time, higher wind speeds result in similar increases in background sound levels (WESTERBERG 1994 in ENGELL-SØRENSEN 2002). Therefore offshore wind turbines sound emissions will vary depending on local conditions and this might cause additional reactions of fish.

Acoustic measurements are time consuming and costly, especially when they are carried out offshore and it is more likely that predicted sound fields in and around offshore wind farms will be calculated. Complex modelling including sound levels, frequency spectra, background noise at different wind speeds, environmental factors such as water depth and sediment type and information about hearing abilities of fish species will be necessary to predict detection and reaction distances for fish. First steps have been taken (DEWI 2004). From the measurement of single turbines (Fig. 1) the sound field of a wind farm and its surroundings was calculated showing the sound levels of different frequency ranges in the surroundings of the wind farm (DEWI 2004). As an example the predicted sound field at an 1/3 octave band of 63 Hz is given in (Fig. 2).

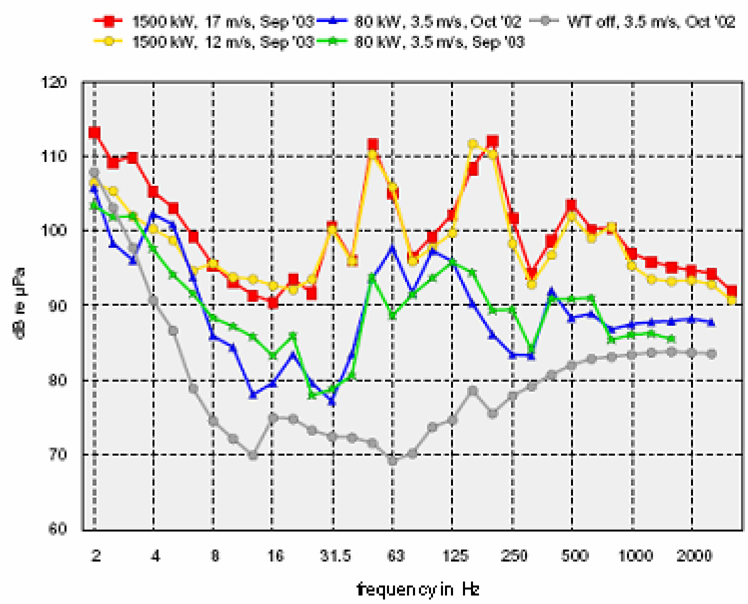


Fig. 1: Underwater sound pressure levels (1/3rd octave spectra) recorded at 110 m distance from the turbine for different turbine states. Wind speeds refer to hub height (nacelle anemometer). The grey line marks the background sound level measured at low wind speeds while the wind turbines were turned off. Figure from DEWI (2004).

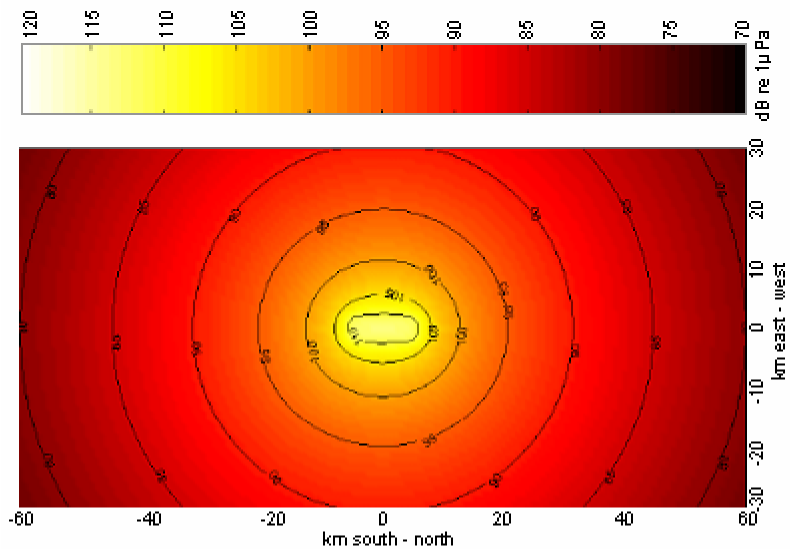


Fig. 2: Underwater sound emission of a hypothetical wind farm containing 70 turbines (5 rows of 14 turbines with a distance of 800 m to each other) calculated on the basis of measurements of a 1.5-MW wind turbine at a medium wind speed. The sound levels are given for an 1/3rd octave band of 63 Hz. Figure from DEWI (2004).

Depending on the background sound level in the sea and other factors such as the foundation material and sediment type, the area in which the sound can be detected varies. And depending on the location of the wind farms the background sound level will not only be influenced by natural conditions such as wind and waves but also by ship traffic and other anthropogenic sound sources. To produce reliable predictions, a strong data base of these factors needs to be build up and more research is necessary.

1.2.3 Hydroacoustics

1.2.3.1 Sound in water and in air

A direct comparison of sound pressure in air and in water in decibels is not possible since, for the same intensity of produced sound, the sound pressure in water is more than 60 dB higher than it is in air. This difference is caused by different sound characteristics in water and air and by different references used for the calculation of decibel in both cases.

The difference in the density of water and air is very large, which causes differences in the speed of sound. The density of air is 1.3 kg m^{-3} the density of water 1030 kg m^{-3}. Sound in air travels with a speed of 340 ms^{-1} while the speed of sound in water reaches 1450 to 1550 ms^{-1} dependent on temperature and salinity (MACLENNAN & SIMMONDS 1992). Caused by the different density and speed of sound in water and air, the sound pressures in air and water differ for sound of equal intensities. The relation between sound pressure in water and air can be calculated from the characteristic acoustic impedance (Z) as followed (VEIT 1979):

$$\frac{P_{Water}}{P_{Air}} = \sqrt{\frac{Z_{Water}}{Z_{Air}}}$$

The characteristic acoustic impedance can be calculated from the density and speed of sound of the medium.

$$\frac{P_{Water}}{P_{Air}} = \sqrt{\frac{1500\,ms^{-1} \times 1030\,kgm^{-3}}{340\,ms^{-1} \times 1.3\,kgm^{-3}}} = \sqrt{3495.5}$$

$$\frac{P_{\ Water}}{P_{\ Air}} = 59.1$$

Transforming this relation into decibel a pressure difference of 20 log 59.1 = 35.43 dB results between sound of equal intensity in water and air.

Additionally the reference for sound pressure in air is 20 μPascal while it is 1 μPascal in water. For this reason the same objective sound pressure in water gives a 26 dB higher sound pressure indication than in air.

$$20 \lg \left(\frac{20}{1}\right) = 26.02 \text{ dB}$$

Therefore the same sound intensity in water yields formally 61.5 dB more sound pressure than in air.

In water, sound waves travel over long distances due to low attenuation. The attenuation increases with frequency and distance.

The wavelength λ depends on the speed of sound and the frequency. In saltwater it can be calculated:

$$\lambda = \frac{1440\ ms}{Frequency}$$

Thus, the length of a 100 Hz wave is 14.4 m in the sea.

1.2.3.2 The acoustical near and far field

An acoustic field can be described in terms of sound pressure and hydrodynamic components such as particle velocity, particle displacement and particle acceleration (VEIT 1979, HAWKINS & MYRBERG JR 1983). In hydroacoustic experiments it is necessary to distinguish between the near and the far field, because of their different acoustic properties. The acoustic situation in a free acoustic field (far field) not restricted by any boundaries is relatively simple. The sound travels in a quasi plane wave and sound pressure p and velocity v are in phase in a ratio of 90°. A sound wave can only be plane in the absence of discontinuities causing refraction and reflection. The hydroacoustic and hydrodynamic components are related to each other and can be easily calculated.

In acoustic near fields such as shallow waters or tanks sound pressure and velocity are out of phase and in the direct vicinity of the sound source the displacement of water causes hydrodynamic effects (VEIT 1979). The hydrodynamic components are of great importance to describe the acoustic field but cannot be calculated from the sound pressure due to complicated reflections and border effects. For this reason the particle movements in near fields need to be measured directly. Measurements of particle movements in water are difficult to carry out (WAHLBERG & WESTERBERG 2005) and are rarely done. Hydrodynamics are of special interest since some fish groups lacking a swimbladder are sensitive to hydrodynamics while most other fish groups with swimbladder can detect sound pressure.

From a monopole sound source the sound waves propagates spherically in all directions. A approximate plane sound wave can only begin at some distance from the sound source because of the growing radius (VEIT 1979). The stronger the spherical wave is curved the stronger is the increase in particle velocity in water caused by a small increase in sound pressure (PARVULESCU 1964). The extension of the near field depends on the wavelength of the sound and is larger at lower frequencies. In the direct vicinity of the sound source the particle velocity is high compared with the particle velocity for the same sound pressure in the far field. In the near field the particle velocity decreases with distance as

$$\frac{1}{r^2}$$

(r = distance from the centre of the monopole sound source.)

while in the acoustical far field it decreases with

$$\frac{1}{r}$$

This latter decrease also holds for the decrease of sound pressure both in near and far fields. The acoustic far field can be defined by the phase equality of velocity and sound pressure (VEIT 1979). But a true far field is an idealized concept (SCHUIJF 1975). Different authors accept different deviations from the ideal far field, which leads to variations in the assumed beginning of the far field. VEIT (1979) defines the far field for distances greater than two wavelengths from the sound source while the figure from HAWKINS (1973) (Fig. 3) gives smaller radii for different frequencies. Conventionally the border between near and far field is considered to be between 1-2 wavelength.

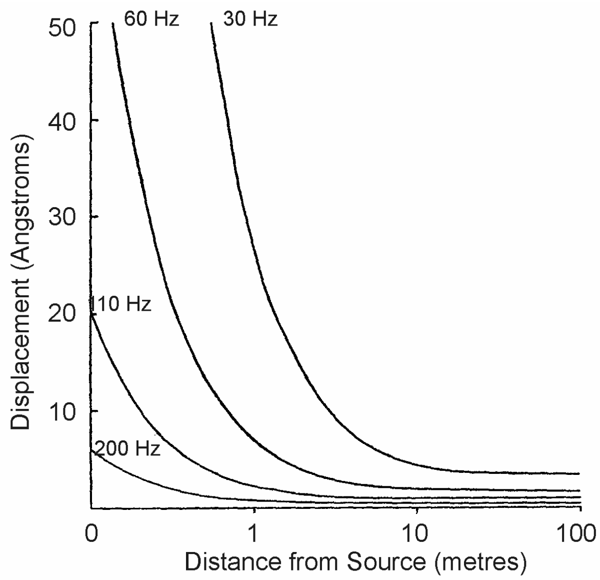


Fig. 3: The “near field” effect, illustrating the decline in the amplitude of particle displacement with distance from a monopole sound source; plotted for a number of different frequencies for a sound pressure of 1 µbar. Figure and legend from HAWKINS (1973).

1.2.3.3 Acoustic in tanks

The acoustic field in experimental tanks is very complicated due to reflections from walls, bottom and water surface. Although the situation in a tank is rather comparable with the conditions of a near field, the near field equations are of only limited use to describe the acoustic field in aquaria.

A number of studies have attempted to minimise reflections at the tank walls by using absorbent materials, such as rubberised hair (TAVOLGA & WODINSKY 1963, BUERKLE 1967). However, at low frequencies relevant to fish, this is very difficult. The material would need to be as thick as about a quarter of the wavelength (FREYTAG 1967), e.g. 3,75 m for sound at 100 Hz.

Due to these complications, research has often been done in special apparatuses, where fish could be placed in well-defined acoustic fields. HAWKINS & MACLENNAN (1975) used a steel tube with sound projectors at each end. Depending on the phase of sound production at the sound projectors the particle motion and sound pressure inside the tube could be varied. The fish were exposed to the sound field in a small cage inside the tube. Another way was to generate the sound in the air surrounding a small thin walled aquarium (FAY 1969). The sound field in air compresses the boundary surfaces of the water mass and sound pressure changes in air lead to sound pressure changes in water with only small particle motion. But this setting is only suitable for small tanks and very low frequencies (HAWKINS & MACLENNAN 1975). Experiments in the wild were done with caged fish that again were confined to a certain sound field (e.g. MCCAULEY et al. 2000).

In contrast, in the present experiments fish were exposed to an inhomogeneous sound field with an obvious sound pressure difference in the tank allowing the fish to avoid highest sound pressure levels.

1.2.3.4 Interferences

In self-contained systems, such as experimental tanks, sound is reflected in different directions and degrees from the walls, bottom and surface. Two sound wave maximum peaks joining will cause an accumulation of sound pressure. If a minimum and a maximum peak join the peaks are levelled out (ATKINS 1973). The sound field in a tank can contain large variations in sound level caused by interferences.

Even under free field conditions, variations in salinity, temperature and pressure can cause reflections and refractions of sound waves, which produce acoustic shadow zones in the sea. Sound travelling over long distances can be changed due to reflections from the surface or the sea bottom (PARVULESCU 1964, HAWKINS & MYRBERG JR 1983). The water surface is an almost perfect reflector due to the impedance differences between the media water and air (HAWKINS & MYRBERG JR 1983).

1.2.4 Fish and sound

1.2.4.1 Hearing ability in fish

Hearing in fish does not generally differ from hearing of terrestrial vertebrates (POPPER 2003). Like other vertebrates fish can discriminate between sounds, determine the direction of a sound source and detect biologically important sounds from background noise (POPPER 2003).

Knowledge of hearing in fish is restricted to about 100 species (HASTINGS & POPPER 2005). The majority of fish are generalists, without specialized hearing abilities and can detect sound up to 500 to 1000 Hz (POPPER 2003). Their best hearing is at frequencies between 100 and 400 Hz, which is also the range of many anthropogenic sounds (POPPER 2003). Hearing specialist can detect frequencies up to 3000 Hz or more with the best hearing ability between 300 Hz and 1000 Hz (POPPER 2003). Some species even detect ultrasound (ASTRUP & MØHL 1993, MANN et al. 2001, GREGORY & CLABBURN 2003).

The inner ear of bony fishes generally consists of three chambers with sensory maculae, the saccule, utricule and lagena, three mechanoreceptor organs connected by semicircular canals (Fig. 4). The macula utriculi is in a mostly horizontal position while macula saccule and macula lagena are vertically postured (HAWKINS & MYRBERG JR 1983).

Every mechanoreceptor organ contains an otolith that covers the epithelial macula, which contains a field of hair cells (Fig. 5). The hair cells in fish have a long kinocilium at the apical end and 40 to 60 shorter stereocilia (depending on species) (POPPER 1978). There are three different types of hair cells that can be found in different areas of the epithelial macula specific to every species. They can be distinguished by the length of kinocilium and stereocilia but the types grade into one another (POPPER 1978).

The otolith is of great importance for the hearing of fish (POPPER 1978). The specific density of a fish is nearly equal to the specific density of water. Sound waves pass through a fish body moving the fish in the same way as the particles of the surrounding water (HAWKINS & MACLENNAN 1975). The higher density of the otoliths compared to fish tissue results in a time delay in their movement, causing shearing forces in the hair cells lying beneath (Hastings & Popper 2005).

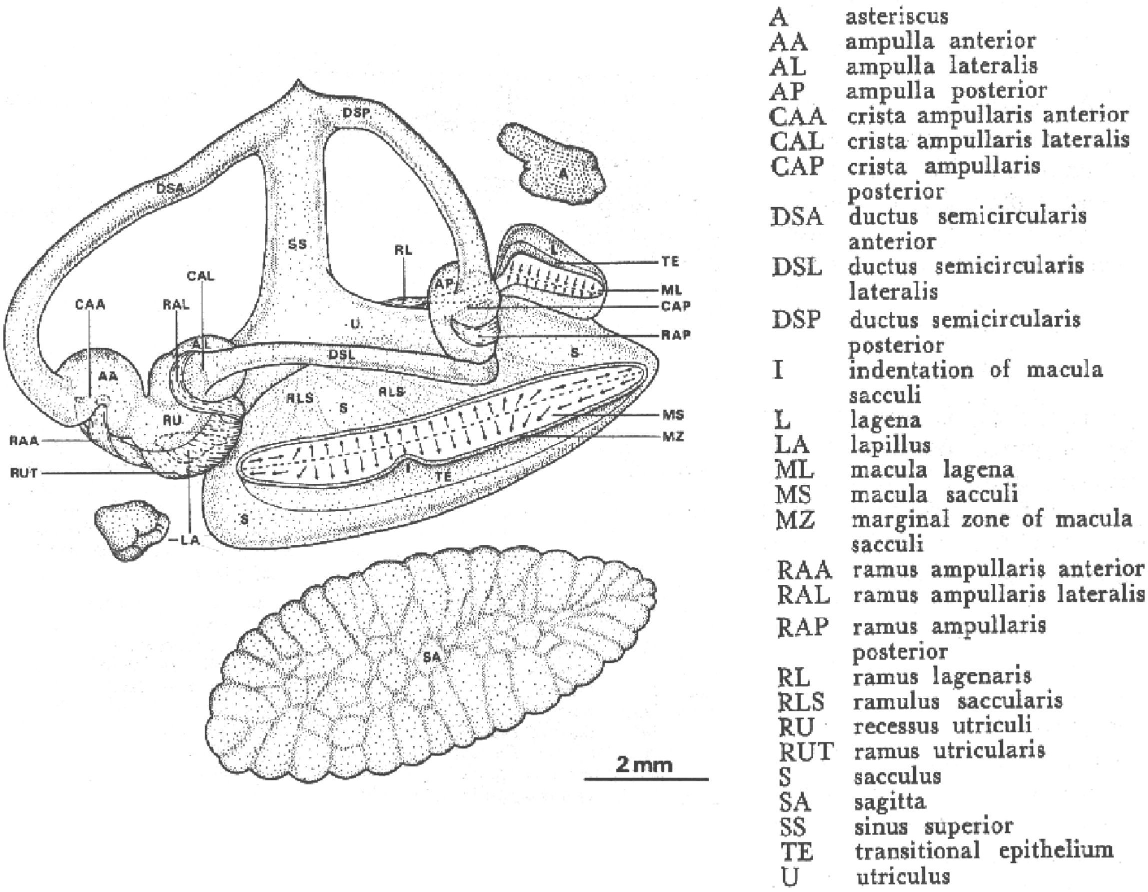


Fig. 4: Left labyrinthine mechanoreceptor organ of cod. (Figure from Dale 1976).

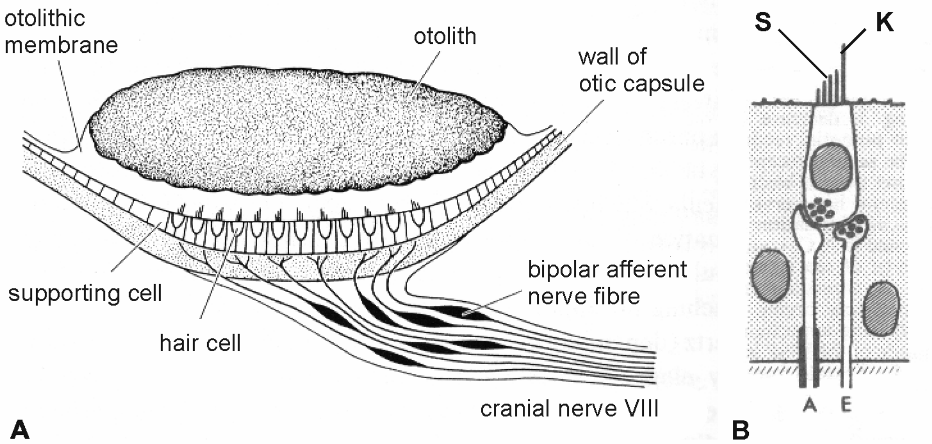


Fig. 5: **A**: Schematic cross section through the utricule of a cod, showing the otolith mounted above the hair cells and separated from them by an otolithic membrane. Figure from HAWKINS (1985). **B**: Hair cell with stereocilia (S), kinocilium (K), afferent (A) and efferent (E) nerve fibres surrounded by supporting cells. Figure after FLOCK &RUSSELL (1973).

The reaction of the hair cells depends on the direction of the otolith movement. Shearing forces towards the kinocilium will cause depolarisation of the cell and stimulation of the afferent nerve. Shearing forces away from the kinocilium will lead to hyperpolarization and an inhibition of the afferent nerve (HAWKINS & MYRBERG JR 1983). The necessary particle displacement to stimulate the hair cells is small and has been determined in cod and two species of pleuronectidae as being from about 0.04 to 0.1 nanometre (CHAPMAN & HAWKINS 1973, CHAPMAN & SAND 1974).

The saccule and lagena are mainly involved in hearing in many teleost fish whilst the utricule is thought to have an equilibrium function (POPPER 1978). However, in some fish species, including clupeids, the utricule also plays a part in hearing, as can sometimes do the lateral line system (HAWKING & MYRBERG JR. 1983).

The hearing threshold of fish can be displayed as an audiogram. It is common practice to produce an audiogram relating to sound pressure or particle displacement. But the important stimulus for the hair cells in the inner ear of fish is the particle acceleration, which is one component of the acoustic wave (SAND & KARLSEN 1986, KARLSEN 1992b, ENGER et al. 1993). Audiograms recalculated from displacement to acceleration show a different picture of the hearing abilities of fish (KARLSEN 1992b). ENGER et al. (1993) calculated hypothetical audiograms relating to sound pressure, particle displacement and particle acceleration that show a clear difference at low frequencies (Fig. 6).

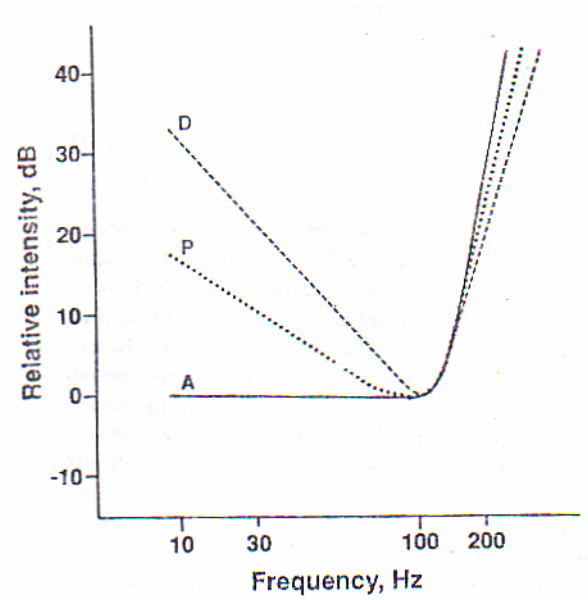


Fig. 6: Hypothetical fish audiograms related to particle displacement (D), sound pressure (P), or particle acceleration (A). Figure from ENGER et al. (1993). In the far field particle displacement, particle acceleration, particle velocity and sound pressure can be calculated using $v = d(2 \pi f)$: $a = d(2 \pi f)^2$. Sound pressure is proportional to v (ENGER et al. 1993).

From the sound pressure and particle displacement audiograms it seems unlikely that fish can detect infrasound, while the acceleration audiogram hints to a good hearing ability for infrasound frequencies. Infrasound may play an important role in hearing in fish since it could enable fish to detect swimming movement of prey and predators that produce predominantly low frequency hydrodynamic noise below 10 Hz (ENGER et al. 1989). KARLSEN (1992b) pointed out that important components of ambient noise are in the infrasound range and SAND & KARLSEN (1986) suggested that detection of infrasound might be utilized for orientation in migrating fish.

The inner ear of fish is sensitive to the particle acceleration of a sound wave. Additionally the sound pressure component of the sound wave can be detected by fish possessing a swimbladder. The swimbladder acts as a pressure-to-displacement transformer (ENGER et al. 1993) converting the sound pressure into particle motion that can be detected by the inner ear. The benefit of the swimbladder as an amplifier does not apply for frequencies below 100 Hz (POPPER 1978, SAND & ENGER 1973). The amount of air in a swimbladder influences the effect on the hearing ability of fish (SAND & ENGER 1973). WAHLBERG & WESTERBERG (2005) pointed out that in fish with small swimbladders that contain only a small amount of air, such as salmonids, the hearing ability is not substantially improved.

The improvement in hearing by the swimbladder also depends on the way the particle displacement is transmitted to the inner ear. In hearing generalists it is transmitted through the surrounding tissue causing attenuation of energy. Therefore, the closer the swimbladder is to the inner ear the better is the hearing ability (POPPER & CARLSON 1998). Hearing ability is enhanced in hearing specialists with different adaptations that improve the coupling between swimbladder and ear (FAY & POPPER 1980). The adaptations in hearing specialists range from extensions of the swimbladder that are very close to the ear (e.g. clupeidae) to direct mechanical connections between the swimbladder and the inner ear as found in ostariophysan fish such as the carp (HASTING & POPPER 2005) that minimizes transmission loss. The sensitivity of hearing specialists extends to a wider frequency range up to 3000-4000 Hz and a hearing threshold that is up to 20 dB lower than the threshold in hearing generalists (HASTINGS & POPPER 2005).

1.2.4.2 Hearing in cod and plaice

Hearing in cod

Cod can detect sound pressure that is converted into particle velocity in the swimbladder. Compared with other hearing generalist cod seems to have a rather narrow hearing range from infrasound as low as 0.1 Hz (SAND & KARLSEN 1986) to about 470 Hz (CHAPMAN & HAWKINS 1973). The upper hearing limit of cod is lower than in most other fish studied. However, while most other teleosts react to a wider frequency range, the hearing ability of cod in its most sensitive range from 60 to 380 Hz is acute and in many cases only limited by ambient noise (SAND & HAWKINS 1973). A reason for low hearing thresholds at the most sensitive frequency range might be the anterior swimbladder projections that extend to the cranium close to the foramen of the ninth and tenth nerves (OFFUT 1974). The labyrinth is located on the other side of this foramen and acoustic stimuli could be transmitted without much loss. These projections are small in juvenile cod but well developed in adults (SAND & HAWKINS 1973) and can also be found in other gadoids (HAGMAN 1921). However in some specimens the projections does not reach the foramen but are tangled next to the swim-bladder (OFFUT 1974, SAND & HAWKINS 1973).

The hearing threshold of cod was determined by various authors (e.g. OFFUT 1974, CHAPMAN & HAWKINS 1973, BUERKLE 1967, 1968) which obtained different results. BUERKLE (1967) estimated a relatively high hearing threshold of about 95 and 100 dB re 1µPa at a frequency range from 20 to 300 Hz which was probably caused by masking of background noise (BUERKLE 1968). CHAPMAN & HAWKINS (1973) found lower hearing thresholds of about 74 to 90 dB re 1µPa and the authors presumed that the threshold have also been masked and should be about 2 dB lower than measured. OFFUT (1974) determined the hearing threshold in a range from 64 to 84 dB re 1µPa. While CHAPMAN & HAWKINS (1973) carried out their experiments in shallow open water, which, it could be argued, provided a better acoustic field and a more natural situation, OFFUT (1974) used an experimental tank. Due to effects of the tank walls, the particle motion might have been higher, which would have caused a stronger stimulus and lower hearing thresholds, since at low frequencies the particle motion is the relevant stimulus for detecting a sound wave with increasing sensitivity at decreasing frequencies (SAND & KARLSEN 1986). Additionally the lateral line can detect particle movement at low frequencies up to a distance of some body length of the fish (SAND & KARLSEN 1986).

OFFUT (1974) did not give any details about the duration and rising speed of the sound stimulus. Quickly rising sounds cause stronger reactions than sound that increases slowly. IVERSEN (1969 in SCHWARZ 1985) pointed out that changes in sound cause stronger behavioural reactions than the maximum sound level and that sudden sounds caused the strongest avoidance.

CHAPMAN & HAWKINS (1973) found a steep rise in the hearing threshold above 400 Hz in cod and conditioning to sound was not possible at frequencies of 520 Hz and above. They concluded that cod is insensitive to high frequency sound. In a range from 60 to 360 Hz the hearing threshold was closely related to the background sound level with a signal to noise ratio of about 16 dB (CHAPMAN & HAWKINS 1973). At frequencies between 60 Hz and 160 Hz the distance from the sound source did not influence the hearing threshold while at the lower frequencies the threshold decreased in the vicinity of the sound source indicating a change of the relevant stimulus from sound pressure to particle displacement (CHAPMAN & HAWKINS 1973). At a frequency range from 60 Hz to 160 Hz (higher frequencies could not be tested) the auditory system of cod seems to be sensitive to sound pressure and the authors expect this to be valid for the higher frequencies in the hearing range too (CHAPMAN & HAWKINS 1973).

Signals of a given frequency are most effectively masked by noise of the same and adjacent frequencies (HAWKINS 1993). To improve the ability to distinguish between a signal and the background noise, cod, like many fish and other vertebrates including humans, possess auditory filters that can be tuned to frequencies of interest (HAWKINS 1993). Inside the frequency range of the filter the hearing threshold is lower than outside the filter range and the bandwidth of a filter varies with frequency (HAWKINS & CHAPMAN 1975).

Cod is capable of distinguishing between the directions of different sound sources (horizontal and vertical direction) (BUWALDA et al. 1983) and between sound sources of different distances (SCHUIJF & HAWKINS 1983). ASTRUP & MØHL (1993) discovered that cod can detect ultrasound of a frequency of 38 kHz and presumed this would enable the fish to detect the echolocating of odontocetes in a range between 10 and 30 meters.

The hearing ability of cod might be different in juvenile and adult fish. ENGÅS et al. (1993) investigated the catch rates of cod and haddock (*Melanogrammus aeglefinus*) before and after seismic shooting, with up to 249 dB re 1 µPa at 1 m distance, and found that the reduction in catches in larger fish was stronger than in smaller fish. This may be related to better hearing ability in adult cod, although another explanation given by the authors was the higher swimming speed of adult cod that allows larger fish to depart the area more quickly whilst smaller fish might habituate to the sound before they are able to leave the affected area.

Sound production in cod

Sound production is widespread in fish and the mechanisms of sound production have evolved independently in different fish group (HAWKINS & MYRBERG Jr 1983). Some gadoids such as the cod produce low frequency grunts using muscles attached to the swimbladder (HAWKINS & RASMUSSEN 1978). BRAWN (1961) looked at *Gadus callarias* and found it to possess similar sound production behaviour. Both sexes produce the same grunts during defensive and aggressive behaviour and when startled, independently from context (BRAWN 1961, HAWKINS & RASMUSSEN 1978). Sound production is more frequent from the end of September to the end of November, due to aggressive behaviour and again in February and March during the spawning season (BRAWN 1961). During spawning season only the male is vocal, using grunts to attract ripe females and to scare away competitors (BRAWN 1961, HAWKINS & RASMUSSEN 1978). The majority of sounds produced by cod are single grunts that can be combined to groups of up to four grunts with about 300 ms from the start of one grunt to the start of the next (HAWKINS & RASMUSSEN 1978). However, cod have also been shown to produce sonic clicks of about 6 kHz at a sound level of 150 dB re 1µPa at 1 m in the presence of two species of seals or human divers (VESTER et al. 2004). The authors supposed the sound to be used as a deterrent against predators. While cod uses a simple repertoire of grunts, haddock uses different grunts and knocks in different social contexts (HAWKINS & RASMUSSEN 1978).

Masking

Masking of biologically relevant sounds by anthropogenic noise is an important issue. Many fish use sound for intraspecific communication in different social contexts. Masking of mating calls could cause serious problems in reproduction. Haddock mating calls were measured at a sound level of 120 dB re 1µPa (A.D. HAWKINS pers. communication) and on the same basis WAHLBERG & WESTERBERG (2005) calculated a detection distance for intraspecific communication of 4 m at a given wind speed of 13 ms^{-1}, although this was based on sound pressure alone and might contain inaccuracies and variations (WAHLBERG & WESTERBERG 2005). This calculation is based on solitary male display sounds but other mating calls are much weaker and more prone to masking (A.D. HAWKINS pers. communication). Spawning in gadoids takes place in the Baltic Sea mainly from February to April (BAGGE et al. in ICES 2005b) and in the North Sea from January to April (BRANDER 1994 in ICES 2005b). It occurs in deeper offshore waters (HAWKINS & AMORIM 2000) under low light conditions that make other senses such as hearing more important. Offshore wind farms are currently restricted to more shallow waters of up to about 25 m and therefore it is unlikely that they would be sited in spawning areas but the sound levels could carry over some kilometres (see chapter 1.2.2) and could mask low level communication. In particular this should be taken into account during the construction phase, which will influence a wider area and should therefore be timed to avoid the spawning season if known spawning grounds are close by.

Hearing in plaice

Plaice is restricted to detection of hydrodynamic components of the sound wave (HAWKINS & MYRBERG JR 1983) due to its lack of a swimbladder. For this reason the hearing ability of plaice is rather poor with a narrow frequency range up to 250 Hz and high hearing thresholds between about 90 and 105 dB re 1µPa (CHAPMAN & SAND 1974). HAWKINS & MACLENNAN (1975) recorded saccular microphonic potentials of plaice in a frequency range from 10 to 250 Hz with highest amplitudes around 90 Hz. They also found that particle velocity, not sound pressure was the stimulus, since changes in sound level did not lead to potential differences but changes in particle velocity did. CHAPMAN & SAND (1974) did experiments on two species of pleuronectidae and obtained comparable results with highest sensitivity in the range between 110 Hz and 160 Hz. KARLSEN (1992a) measured a sensitivity of plaice to infrasound as low as 0.1 Hz using a cardiac conditioning technique. The audiogram of plaice showing the results of KARLSEN (1992a) and the recalculated results of CHAPMAN & SAND (1974) is displayed in Fig. 7. This audiogram gives a sensitivity of plaice to frequencies from infrasound to a frequency of about 200 Hz.

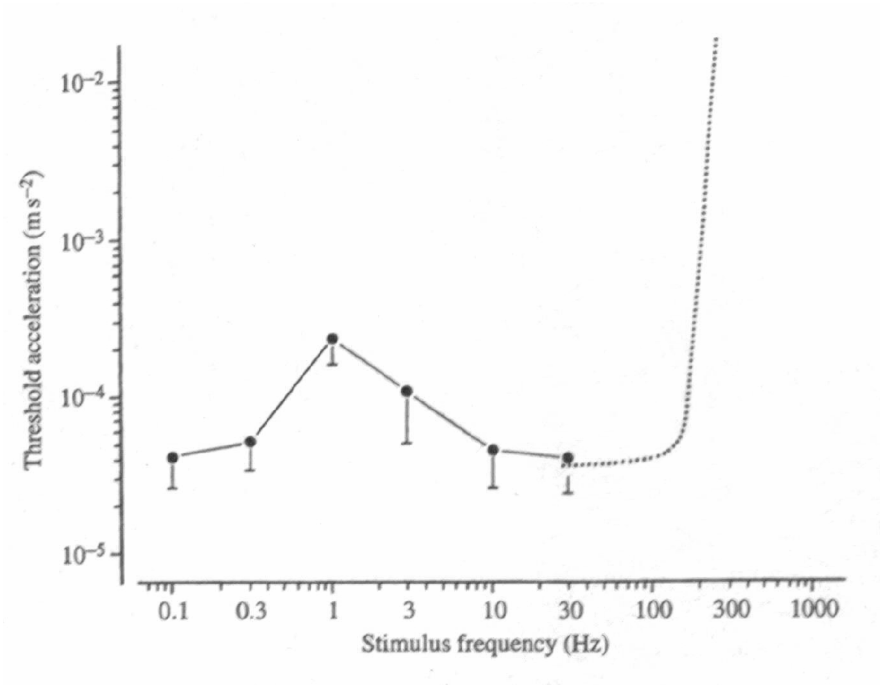


Fig. 7: Auditory thresholds obtained in six plaice for the frequency range 0.1-30 Hz, presented as mean values + S.D. The dotted curve give the acceleration thresholds found in plaice by CHAPMAN & SAND (1974). Figure and legend from KARLSEN (1992a). Elevated thresholds at 1 Hz and 3 Hz might be caused by masking.

1.2.4.3 Reaction of fish to anthropogenic noise

It is well established that fish can hear, but knowledge of the influence of man-made noise on fish is still small (e.g. MCCAULEY et al. 2003, POPPER 2003, AMOSER et al. 2004, POPPER et al. 2005, WAHLBERG & WESTERBERG 2005). Many fish are sensitive to a wide range of frequencies and sound production for communication is widespread (HAWKINS 1973, SAND & KARLSEN 1986, ASTRUP & MØHL 1993, POPPER 2003). The acoustic environment can be defined as the portion of the sound spectrum in water to which a fish is sensitive (MOULTON 1963). Most anthropogenic noise is within the hearing range of fish, and may affect behaviour and/or physiology and might cause either temporary or permanent effects (POPPER 2003). Anthropogenic noise may impair fish hearing leading to difficulties in orientating in the acoustic environment, finding prey, avoiding predators and communication (MCCAULEY et al. 2003, POPPER 2003). Anthropogenic sound can be caused by stationary sound sources such as oilrigs, coastal plants and offshore wind farms or by temporary sources such as ships (e.g. AMOSER et al. 2004, VABØ et al. 2002), seismic surveys (e.g. MCCAULEY et al. 2003) and pile driving (e.g. NEDWELL et al. 2003b) that can cause high sound level peaks.

To evaluate the effects of anthropogenic noise on fish it is necessary to know how fish react to sound and to determine thresholds that lead to temporary or permanent hearing loss or other effects that can impact the survival of individual fish or fish stocks (POPPER 2003).

One problem is masking of important sounds by anthropogenic noise (POPPER 2003). Masking is the inability to separate a signal from background noise (HAWKINS & CHAPMAN 1975). Fish with low hearing thresholds are particularly affected by noise masking signals. Signal detection depends not only on the sound level of the signal and the hearing ability of the fish but on the relation between signal and background noise (signal-noise-ratio) and in a noisy environment like the sea, the ability to discriminate between a signal and the background noise is more important than the absolute hearing threshold (HAWKINS & CHAPMAN 1975). CHAPMAN & HAWKINS (1973) suggested that ambient noise in the sea limits the hearing abilities of the majority of marine fish especially under adverse sea conditions. LEIS et al. (2003) revealed that the larvae of some reef fish species use reef sounds for orientation during settlement. Masking of reef sound by noise from offshore could cause confusion leading the larvae away from the reef and preventing their settlement in the reef (POPPER 2003). WESTERBERG (2000) mentioned possible masking of communication sounds by offshore wind farm noise during spawning as an important subject that needs to be investigated. Some schooling fish, such as herring (*Clupea harengus*) and sprat (*Sprattus sprattus*) use low frequency sound detected by the lateral line organ to maintain their school structure and could be affected by masking of this sound (KRAAN & VAN ETTEN 1995).

Fish can react to noise in different ways and the response might depend on life stage, physiological state (POPPER 2003) as well as diel rhythms, temperature and season (NEPROSHIN 1979). MISUND & AGLEN (1992) described avoidance behaviour of herring and sprat to low frequency vessel noise and presumed that the strength of the reaction depended on the distance of the sound source when first detected by the fish. Noise approaching slowly from greater distance would cause smaller reactions than noise appearing suddenly at short distance. SCHWARZ (1985) presumed that most fish would ignore even high level continuous sound if it is not combined with other stimuli or connected with danger or a reward. This was based on observations such as IVERSEN'S (1969 in SCHWARZ 1985) that skipjack tuna (*Katsuwonus pelamis*) swam next to a noisy ship until the speed and therefore the sound level suddenly changed. After the sound stabilized on the new level the fish returned to the boat. The same effect was observed in Atlantic herring (*Clupea harengus harengus*) (HERING 1968, OLSEN 1970 in SCHWARZ 1985).

NEPROSHIN (1979) found a diel rhythm in the reaction distance of pacific mackerel to vessel noise depending not only on the noise but on light, temperature, feeding activity and season. ENGÅS et al. (1996) described significantly lower catch rates of cod and haddock during and after the use of seismic guns but it is not known whether the fish were scared away, injured or even killed and whether increasing fish numbers afterwards were caused by the same fish returning or by other fish replacing the reduced stock. Startle and alarm responses in rockfish during exposure to air-gun sound were revealed by PEARSON et al. (1992). WARDLE et al. (2001) observed startle response to single air gun shots but the fish would only move away from the sound source when an additional visual clue indicated the direction of the sound source. The authors therefore presumed that the single shot of an air gun was too short or too complicated in its composition to provide directional information to the fish. Additionally loud noise might cause fish to “freeze on the spot“, exposing it to high sound levels that might cause physiological damage (POPPER 2003).

MCCAULEY et al. (2000) observed the behaviour of pink snapper (*Chrysophrys auratus*) in a net cage during air-gun shots. While the fish disappeared from view and dived to the bottom of the cage after the first shot they stayed closer to the camera at the second shot although moving to lower areas of the cage. The same fish were exposed to three more air gun shots after recovery time of 58 days but did not show any obvious reaction to the sound. Whether the lack of behavioural reaction was caused by habituation to the sound or deafness of the fish is not known but damage to the hair cells was evident (MCCAULEY et al. 2000). Sound might cause slight short-term effects but it might also cause flight reactions scaring fish from feeding grounds or spawning areas, which could have permanent effects on fish stocks (POPPER 2003). HANDEGARD et al. (2003) measured individual responses of fish using an acoustic target tracking method. The authors showed vertical and horizontal movements of individual cod when approached by a trawling vessel. This work was extended to investigate the reactions to different types of sounds related to trawling (HANDEGARD & TJØSTHEIM 2005).

CHAPMAN et al. (1974) described attraction of some fish species including cod to low frequency sound produced by breathing divers. In this experiment fish showed avoidance behaviour to the divers but became used to the sound probably while taking advantage of additional food from the disturbed sea bed during diving activity. In later playback experi-ments the fish were attracted to the sound of the divers. CHAPMAN (1976) observed attraction of juvenile gadoids to pulsed low frequency sound which might be related to natural food searching behaviour with pulsed tones resembling the sound of fish movements. MOORHOUSE (1932) observed rapid habituation of perch to sound in a tank and WAHLBERG & WESTERBERG (2005) mentioned that it would be relatively easy for a fish to associate a sound with certain events and habituation to a sound that is not connected with danger could occur. However, it is not known how fish might react to a long lasting continuous sound.

1.2.4.4 Results from offshore wind farm research

There are only a few studies investigating the effects of offshore wind turbine sound emissions on fish. In addition, a number of studies have examined the influence of the sound produced during pile driving, which is of importance during construction of offshore wind farms (e.g. McKenzie Maxon 2000, Nedwell et al. 2003a).

The sound of pile driving is on a higher level than the operational sound of wind turbines, but the sound emission is not continuous. The sound signal will depend on the material of the pile, the sediment structure and the way the piling takes place and efforts are made to monitor and reduce sound emissions (Vagle 2003, DEWI 2004). Measurement during pile driving in the Baltic Sea showed sound levels of about 175 dB re 1µPa declining to about 159 dB re 1µPa at a distance of 1100 m (DEWI 2004). Nedwell et al. (2003a) measured source levels of 260 dB re 1µPa at a distance of 1 m during piling and calculated a sound level of more than 170 dB re 1µPa at a distance of 10 km from the turbine. A study carried out by Feist et al. (1992 in Hawkins 2003) showed that the number of juvenile salmonid schools was reduced to about one half during pile driving compared with the same area when pile driving was not taking place, indicating that the fish returned after the noise stopped. The authors estimated a detection range for the hearing generalists of at least 600 m at a source level of 150 dB re 1µPa.

The possible influence on fish of the noise and electromagnetic fields created by the world's first commercial offshore wind farm, Vindeby (Denmark, 11 turbines of 450-kW established in 1991) was evaluated by Engell-Sørensen (2002) after ten years of operation. The evaluation was prompted by anecdotal evidence provided by the only commercial fisherman active in the area, that catches of the flatfish turbot (*Psetta maxima*) decreased inside the wind farm when wind speeds were higher than 5 ms^{-1} compared to catches outside the wind farm. The fisherman also observed that 24 hours calm weather conditions were needed before turbot would return to the wind farm area. Planned catch experiments to confirm or reject these findings had to be cancelled due to late arrival of turbot in spring and bad weather conditions. Instead, the author compared data in the literature on the hearing ability of flatfish with the Vindeby sound emissions of 85 to 120 dB re 1µPa in a frequency range from 0,1 Hz to 400 Hz measured at a distance of 14 m from the turbine. The study concluded that an avoidance reaction of turbot and flounder (*Psetta flesus*) was possible but not likely, even when the fish were close to the turbine during high wind speeds.

HAUMANN (1993 in ENGELL-SØRENSEN 2002) examined the Vindeby wind farm area before construction and during operation and found a considerable increase in the number of cod in the area of wind farm, which the author attributed to the artificial reef effect. Apart from this, HAUMANN concluded that noise or other physical impacts caused by the wind farm did not have any negative effects on fish or fishing. However, as pointed out by ENGELL-SØRENSEN (2002) these conclusions were not based on statistical analysis.

The strength of an artificial reef effect depends not only on the structure of the artificial substrate but also on the surrounding environment since recruitment is subject to the organisms in surrounding areas and depends on currents carrying larvae and spat (ENGELL-SØRENSEN et al. 2000). JOSCHKO et al. (2004) expected turbine piles to change the hydro-dynamic regime and the sedimentary environment of the soft bottom, which could cause changes in larval settlement. Additionally the piles would represent an artificial hard substrate introduced into a soft bottom area, which would be likely influence the ecosystem significantly by allowing certain species, such as cirripedia, actinaria and bivalvia to colonize the area (JOSCHKO et al. 2004). From research in the wind farm area Horns Rev an 8-fold increase in food availability (epifauna-biomass) for fish was estimated due to introduction of hard substrate, which could result in an increase in fish abundance (ELSAM ENGINEERING 2004). Hydroacoustic fish monitoring was initiated in 2004 and indicated that the wind farm attracted fish beyond a distance of 500 m (HVIDT et al. 2005). An average increase of 300% in sandeel abundance was recorded compared to the year before the wind farm was erected, although abundance decreased by about 20% in the control area outside the wind farm area (ELSAM ENGINEERING & ENERGI E2 A/S 2005).

WESTERBERG (2000) observed increasing fish numbers in an area of less than 200 m and lower catch rates at a distance between 200 m and 800 m around the 220-kW Svante wind turbine (Sweden) when the turbine was switched off compared to when it was operational. The sound level emitted by the small turbine was 102 to 113 dB re 1µPa at a distance of 1 m depending on the wind force (WESTERBERG 2000). A possible explanation given by the author was an artificial reef effect caused by the pile and that attraction and avoidance were balanced during operation of the turbine. Therefore the fish moved closer to the attractive pile when the turbine was switched off while they moved to the less noisy area during operation.

VALDEMARSEN (1979) observed significant increases in catches of saithe (*Pollachius virens*) and cod in the vicinity of an oilrig compared with catches 500 m away from the platform and presumed that demersal fish would aggregate around oilrigs. Flatfish such as plaice can also be attracted to artificial reef structures (POLOVINA & SAKAI 1989 in HOFFMANN et al. 2000).

The environmental monitoring programme at the offshore wind farm Horns Rev containing 80 turbines of 2-MW raised the subject of sound influence on fish (ELSAM ENGINEERING & ENERGI E2 A/S 2005), but did not include research on changes in fish communities due to construction and operation of the farm. The authors expect the sound produced from offshore wind turbines to be different from natural sound sources including other marine organisms and therefore masking of biological sounds would not be a problem. Additionally continuous sound production would be likely to result in habituation of fish.

The report of ENGINEERING & ENERGI E2 A/S (2005) mentioned that before the wind farm was erected the Horns Rev area was a nursery area for herring with larvae present in spring and juveniles present in the summer and autumn.

The area of the Nysted wind farm containing 72 turbines of 2.3-MW is presumed to be part of a large feeding, breeding and spawning ground used by a number of fish including cod, haddock and herring (BIO/CONSULT 2000 in ELSAM ENGINEERING & ENERGI E2 A/S 2005). Masking of vocal species such as cod and haddock could negatively effect spawning success.

The environmental statement for the offshore wind farm Robin Rigg (NATURAL POWER 2002) expects the sound emission even during pile driving to be too low to affect fish but the estimation was based on a high reaction threshold of 180 dB re 1µPa even for hearing specialists. HAWKINS (2003) criticized the statement given by NATURAL POWER (2003) since it would underestimate the effect of pile driving which might be significant and can even kill fish. The author measured pile-driving noise in levels detectable for sound sensitive fish such as cod, shad (*Alosa spec.*) and herring at a distance of 20 km away from the sound source.

A possible negative effect of offshore wind farms could be changes to the electro-magnetic field in the direct vicinity of the cables. Research on this subject is rare and therefore it is not known if fish species sensitive to electro-magnetic fields such as sharks, rays and some bony fish might react to electro-magnetic fields produced by offshore wind farms (ENGELL-SØRENSEN 2002) A study carried out on dogfish (*Scyliorhinus canicula)* showed some avoidance to electric fields at 1000µV/m but with high variability among individuals (GILL & TAYLOR 2001)

All these studies highlight the lack of in depth field studies and information for key species and life history stages, such as vocalizing adults during spawning season, larval stages etc. Standardisation of research projects and long term monitoring before, during and after wind farm construction would be highly desirable (BSH 2001).

1.2.4.5 Hearing threshold shift and inner ear damage

Little is known about the effect of noise on the inner ear of fish (HASTINGS et al. 1996). A number of studies examined the influence of intense noise on the ear of fish (e.g. POPPER 1974, 1978, 2003, 2005, ENGER 1981, HASTINGS et al. 1996, SCHOLIK & YAN 2001, 2002a,b, MCCAULEY et al. 2003, SMITH et al. 2004). In some species, temporary hearing threshold shifts could be seen that lasted from a few hours to several days depending on fish species, duration and frequency of the produced sound (SCHOLIK & YAN 2001). Experiments on goldfish (*Carassius auratus*) showed a 5 dB threshold shift after 10 minutes of sound exposure (0.1-10 kHz, 170 dB re 1µPa) increasing linearly to about 28 dB after 24 hours with no further increase with longer durations of exposure (SMITH et al. 2004). More than two weeks were necessary for the hearing threshold to get back to the normal level after sound exposure of 21 days (SMITH et al. 2004). Fathead minnows (*Pimephales promelas*) were exposed to sound for 24 hours and not all the fish tested recovered during the following two weeks (SCHOLIK & YAN 2001). Temporary (or permanent) hearing loss seems to depend on the hearing threshold of fish and hearing specialists (such as goldfish and fathead minnow) can be affected by sound levels that do not show any significant effect in hearing generalists (SCHOLIK & YAN 2002a, POPPER 2003).

In addition to temporary threshold shifts, damage of the hair cells in the inner ear of fish can occur. MCCAULEY et al. (2003) held pink snapper (*Pagrus auratus*) in cages in a shallow bay, exposed them to air-gun noise and examined the ears of some individuals (Group I) before sound exposure and others 18 hours (Group II) or 58 days (Group III) after exposure. The peak-to-peak air-gun source level was 222.6 dB re 1µPa at 1 m distance. At 100 m the sound level was more than 25 dB above background noise. The main energy was in a range from 20 to 100 Hz, with main energy between 100 and 1000 Hz, which is within the range of peak sensitivity of many fish. The results showed significant damage in the hair cells of group III compared with the samples taken from group I and II. There was no indication of regeneration of hair cells in the damaged epithelia within 58 days after exposure. In these experiments the fish could not escape the intense sound that caused damage of the inner ear. In the open sea it is possible for the fish to avoid highest continuous sound levels by swimming away from the sound source as the fish in the cages tried to do.

Enger (1981) exposed cod to intense sound of different frequencies within a range from 50 to 400 Hz at a sound level of 180 dB re 1µPa which is about 100 to 110 dB above the hearing threshold in the most sensitive hearing range of cod. The duration of the sound was between 1 and 5 hours and the fishes were sacrificed after exposure. ENGER (1981) found areas with a complete or almost complete lack of hair cells in different regions of the saccular macula depending on frequency. While the central area of the macula saccule was affected in all tested frequencies, higher frequencies seemed to cause damage in the anterior region and lower frequencies seemed to affect the posterior portions of the macula. Besides evidence for a coarse frequency discrimination at the macula saccule the results show obvious damage of the inner ear immediately after sound exposure as also shown in MCCAULEY et al. 2000). In other experiments (HASTINGS et al. 1996, MCCAULEY 2003) significant damage became obvious only after some time after exposure and increased with time. However, even if damage is not obvious, the function of hair cells might be affected (MCCAULEY et al. 2000).

While in humans, all hair cells are produced before birth and losses cannot be compensated by regeneration, fishes, amphibians and birds can proliferate hair cells during growth and might be able to regenerate hair cells throughout their lives (POPPER & HOXTER 1984, CORWIN & OBERHOLTZER 1997). It has been shown that the hair cells of the inner ear of fish regenerate after damage with chemicals (LOMBARTE et al. 1993) but it has not so far been shown that regeneration will occur after exposure to intense sound.

POPPER & HOXTER (1984) found that the number of hair cells increased in the macula saccule of *Astronotus ocellatus*, corresponding to body size up to a certain length, above which numbers remained constant. The higher number of hair cells might be related to increasing hearing abilities in older fish. While POPPER (1971) did not find any difference in hearing ability in two groups of goldfish (*Carassius auratus*) of different body length, KENYON (1996) found the hearing ability in two species of bicolour damselfish (*Pomacentrus partitus* and *P. variablis*) increased with size and age of the fish.

The hair cells of the lateral line, which are the same type as in the inner ear, could be affected by sound in the same way as the inner ear but this has not been addressed in studies yet (POPPER 2003).

1.2.5 Biology of plaice and cod

1.2.5.1 Biology of cod

There are different geographic stocks of cod in Atlantic, North and Baltic Sea. The North and Baltic Sea stock contains about 10% of the overall population and is independent from the Atlantic cod stock (VORBERG & BRECKLING 1999). Cod mostly live close to the bottom down to some hundreds of meters, but juveniles are frequently found in river mouths (VORBERG & BRECKLING 1999). Juveniles feed mostly on crustaceans and sometimes polychaetes, adults prey on crustaceans, herring, sandeel and other fish (VORBERG & BRECKLING 1999). In daytime cod can aggregate to large schools (VORBERG & BRECKLING 1999).

Spawning time in the North Sea is from January to May. After a planktonic period juvenile cod settle near the bottom at an age of between three to five months and stay there for two years (VORBERG & BRECKLING 1999). Some cod reach maturity in their second year but most cod mature in the age from 3 to 4 years (Fig. 8).

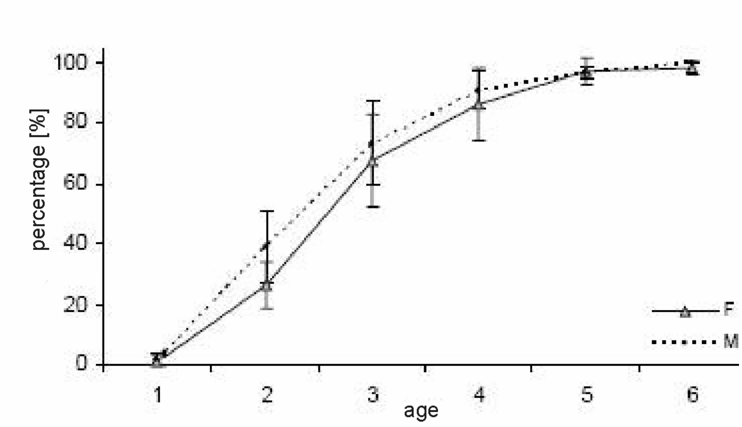


Fig. 8: Percentage of mature fish in relation to age in male and female cod (data of International Bottom Trawl Survey 2000-2004). Figure from ICES (2005c).

In the North Sea the population of cod is dominated by 1-year- (10-25 cm) and 2-year-old (20-40 cm) juveniles (ICES 2005c). Adult cod in the North Sea migrate between summer feeding grounds in the north and southern spawning areas in winter. Spawning grounds are widespread over the North Sea (ICES 2005c). GODØ & MICHALSEN (1997) observed vertical migration in cod by data storage tags but could not see a diurnal rhythm in the behaviour.

1.2.5.2 Biology of plaice

The distribution of plaice extends from the North and Baltic Seas, along the East Atlantic coast to western parts of the Mediterranean Sea (VORBERG & BRECKLING 1999).

Plaice is a bottom-living fish that feeds mostly during daytime (VERHEIJEN & DE GROOT 1967) on benthic invertebrates including polychaetes, amphipods and mussels (RIJNSDORP & VINGERHOED 2001). Juvenile plaice live near the shore and move into deeper water as they grow older (HEINCKE 1913). Maturation occurs in males between 2 – 4 years old (20 - 30 cm length), and in females at 4 – 5 years (30 - 35 cm length) (RIJNSDORP 1989) (Fig. 9).

Spawning takes place in spawning grounds in the central and southern North Sea (ICES 2005b) at water depths between 20 and 40 m between January and April (VORBERG & BRECKLING 1999). The eggs and larvae are planktonic and appear near the shore mostly on the eastern side of the North Sea in spring (ICES 2005b). At a size of 10 to 12 mm the pelagic larvae metamorphose during which the left eye moves onto the right side of the body and the fish turns the left side to the bottom (VORBERG & BRECKLING 1999).

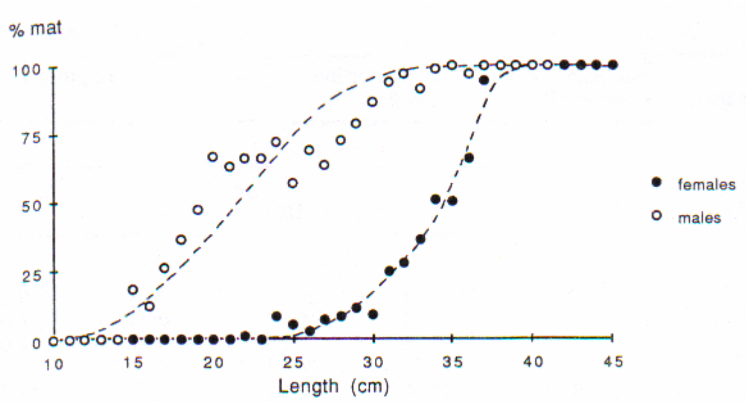


Fig. 9: Percentage of mature fish in relation to length in male and female plaice (data of surveys in 1985 and 1986). Figure from Rijnsdorp (1989).

GIBSON (1980) reported the behavioural repertoire of juvenile plaice and described the following behaviour as common:

- Resting at the bottom
- Resting buried in sediment
- Slow swimming at the bottom ("shuffles", "shambles")
- Fast swimming elevated off the bottom
- Little movements on the spot or without moving more than half of the body length ("shift")

Shorter behavioural actions such as biting, yawning, U-bend and roll movements were reported (GIBSON 1980). ARNOLD (1969) observed that plaice tended to turn their head into the direction of water flow. Given the hydrodynamic shape of the plaice body this orientation prevents water from flowing underneath the body.

VERHEIJEN & DE GROOT (1967) and GIBSON et al. (1978) described higher activity of plaice at the bottom of tanks during daytime whilst fish swam higher off the bottom at night. VERHEIJEN & DE GROOT (1967) related the results to lower catch rates in bottom trawls during night tows as the trawl may pass underneath the swimming fish. The higher activity in daytime was thought to be explained by feeding activity at the bottom, although no explanation could be given for the behaviour at night (VERHEIJEN & DE GROOT 1967). The activity of juvenile wild plaice is connected to the tides with two activity peaks when they inhabit beaches with obvious tides (GIBSON et al. 1978). The activity of adult plaice seems to be controlled by an endogenous circadian rhythm with a tendency to higher activity at the beginning and end of the light period (if light was present). This rhythm was observed with slight differences in day/night light conditions as well as in constant light and constant darkness (GIBSON et al. 1978).

1.2.5.3 Stocks of cod and plaice in the North Sea

Like many other species in the North Sea, the size of cod and plaice stocks has declined over the last few decades (ICES 2005b). Both stocks are outside safe biological limits (FRID et al. 2003) and are considered to be at risk because of reduced reproductive capacity (ICES 2004). The minimum landing size of plaice is 27 cm but many undersized plaice are caught and discarded as bycatch in sole fishery (ICES 2005b). In 2004 the spawning stock biomass (SSB) of plaice in the North Sea was estimated at around 170,000 t and at just above 200,000 t in 2005 which is just below the precautionary biomass (B_{pa}) of 230,000 t. The limit biomass (B_{lim}) was defined at 160,000 t (ICES 2005a).

The minimal landing size of cod is 35 cm in North Sea and 33 cm in the Baltic Sea (VORBERG & BRECKLING 1999). Given the minimum landing size for other species such as plaice and haddock is smaller than cod, the mesh sizes used in many commercial fisheries result in immature cod being caught before they have a chance to reproduce. In addition the central Baltic Sea stock is at a historical low point (ZIMMERMANN & GRÖHSLER 2004). Presently the spawning stock biomass of cod in the North Sea is well below the defined precautionary biomass of 150,000 t and even below the biomass limit of 70,000 t (ICES 2005a).

2 Issue of the investigation

The aim of the project was to examine the reactions of cod and plaice to constant low frequency sound resembling offshore wind turbine noise in frequency range and sound levels. Different frequencies were tested to identify those that might cause greater effects in cod or plaice. This was important in order to estimate the influence of offshore wind farms on fish populations, to adapt the turbines with regard to sound emission if necessary and to estimate the detection and reaction distance of fish species in the vicinity of a sound source.

A sound pressure difference in the tank was produced using Styrofoam sound barriers to divide the tank into connected quarters. These sound barriers used the border effect between water and air (contained in the Styrofoam) to reflect the sound within the tank quarter of sound production.

Cod and plaice were chosen as experimental fish. Both are commercially important species, but with different hearing abilities and behaviour that might be affected by the sound of offshore wind turbine farms. The low stocks of several fish species in North and Baltic Seas including cod and plaice make it necessary to identify and minimize potential risks of anthropogenic sound sources such as offshore wind farms for fish. Two different age groups of cod and plaice were tested since it is possible that the reaction of fish to sound varies during development.

The fish in the tank experiments were exposed to sound for 24 hours periods, which allowed observation of both the potential startle response but also possible habituation to the sound stimulus.

In summary the aim of the experiments was to answer the following questions

- Can the acoustic field in the experimental tank be controlled/manipulated and the sound pressure difference increased by sound barriers?
- How will cod and plaice react to continuous sound of different frequencies and sound levels?
- Will the sound stimulus be strong enough to expel the fish from a preferred area?
- Can differences in sound-related behaviour of juvenile and adult cod and plaice be seen?
- Are the behavioural reactions of cod and plaice frequency dependent?
- Can the reaction of cod and plaice be related to their hearing abilities?
- Do the fish show habituation to the sound?

3 Material and Methods

The experimental investigations were carried out at the *Fisheries Research Services (FRS) Marine Laboratory* in Aberdeen, Scotland, from February to December 2004.

3.1 Experimental fish

3.1.1 Cod (Gadus morhua)

The groups consisted of 15 juveniles (from which one died) and 13 adults. Their size is given in Table 1. Cod were hard to catch and therefore were sourced on a number of different occasions. 18 cod were caught using rod and line off the Scottish coast near Aberdeen. Three adult cod were caught by trawl further offshore and seven adults which had been caught by trawl and kept in a holding tank for about two years were used to make up the numbers required for the experiments.

Table 1: Total length of juvenile and adult cod measured at the end of the experiments

	number of cod	
TL [cm]	juvenile	adult
32.0	1	
35.0	1	
38.5	1	
41.0	2	
42.0	1	
43.0	1	
46.0	1	
47.0	1	
49.0	2	
51.0	2	
53.0	1	
56.0		1
60.0		1
62.0		1
63.0		1
65.0		1
66.0		2
68.0		1
69.0		1
71.0		2
72.0		2
total number	14	13

3.1.2 Plaice (Pleuronectes platessa)

Plaice were used that had been caught using a small scientific sampling trawl off the Scottish coast near Aberdeen in September 2003. Two different age / size groups were used. One group contained 20 juvenile plaice with a total length ranging from 23.5 to 32 cm. The other contained 20 adult plaice with a total length ranging between 26 and 43 cm. Some of the adults showed an abdominal swelling indicating development of eggs/onset of maturity. During the course of the experimental period three juveniles and two adult plaice died for unknown reason. The final numbers were 17 juveniles, and 18 adults. Their size at the end of the experiments is shown in Table 2.

Table 2: Total length of juvenile and adult plaice measured at the end of the experiments

	number of plaice	
TL [cm]	juvenile	adult
23.5	1	
24.0	3	
24.5	1	
25.0	2	
26.0	2	1
27.0	1	
28.0	3	
28.5	1	
29.0	1	
31.0	1	
32.0	1	2
32.5		3
33.5		1
34.0		1
34.5		2
35.0		4
38.0		1
39.0		2
43.0		1
total number	17	18

3.2 Experimental tank

The experiments were carried out in an annular concrete tank of 10 m in diameter (Fig. 10). It was built in 1970 for behavioural studies on fishes. Low light conditions, reduction of background noise and insulation against vibration serve to increase the well being of fish in the tank. In it's centre, the tank contains an observation chamber of 2.4 m in diameter with eight windows into the aquarium. It can be entered via a moveable gantry above the tank. The annular radius of the tank is 3.68 m. Water depth was 1.26 m, the maximum depth for the tank. The capacity of the tank is about 85,000 l (WARDLE & ANTHONY 1973). Seawater is supplied from a nearby bay. It is filtered through sand, irradiated by UV-light and brought to a constant temperature of 10°C.

All tanks in the aquarium facility are equipped with a joint filter system including biological cleaning, pressure sand filter, coral sand filter, UV-irradiation and protein skimmer. Additionally a small amount of ozone is used for disinfections. The tank contains two water inflows and two outflows. Both inflows and one of the outflows are located near the bottom of the tank. The second outflow being a tube extension ending just beneath the water surface determines the water level. Currents of 0.1 ms^{-1} to 0.12 ms^{-1} and 0.19 ms^{-1} to 0.26 ms^{-1} were measured at the two inflows.

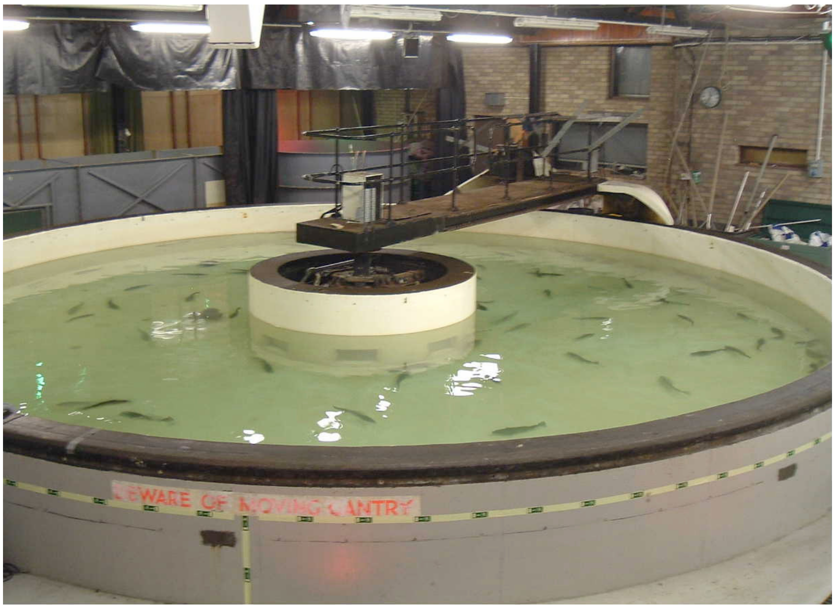


Fig. 10: The experimental tank with some fish, but without barriers in place.

Before the experiments the fish were kept in 3 m holding tanks, 1 m deep, where they were fed three mornings a week. While being in the holding tank the fish were exposed to a green light / darkness regime resembling the natural light conditions depending on the time of the year.

At the start of the experiments the fish were released evenly distributed into the tank allowing seven days for acclimatisation before starting the first experiment. A light regime of 12 hours each of green and red light during daytime and red light at night was set for the experiments. The red light was necessary to enable video surveillance at night and was chosen, as it is believed to be outside the visible range of plaice (GIBSON 1973) and cod (BOWMAKER 1990). Additionally two smaller red lights were installed on both sides of the gantry to lighten up dark areas below the gantry. During the experimental series the fish were fed every second day in the early afternoon including 24 hours before turning the sound on and three hours before turning the sound off. The feed was distributed evenly in the tank. Plaice were fed with fresh fish, cod with fish pellets. During feeding time the state of the fish was checked.

The tank bottom could not be covered with sediment, which would have been more natural conditions for plaice, since the fish buried in sand would be invisible in video observation.

3.3 Experimental settings

In order to create a large sound pressure difference, the tank was divided into four equally sized quarters by sound barriers (see chapter. 3.3.1). The barriers extended from the observation chamber to the periphery. Three of them were shorter than the tank radius leaving a gap of 0.7 m at the outside to allow the fish to move around freely between quarters. The fourth barrier span the annular radius from the central to the outer wall to avoid sound circulating in the tank. Its outer end was placed between the two inflows. The quarters were numbered 1-4 anticlockwise, with 1 and 4 being adjacent to the long barrier. The numerical order of the quarters is indicated in Fig. 11.

For sound production two loudspeakers (chapter 3.3.2) were installed in the quarters 1 and 4 (Fig. 12B). Additionally, two loudspeaker dummies, approximating the loudspeaker size and made from plastic (Fig. 12A) were positioned in quarter 2 and 3. Loudspeaker and loudspeaker dummies were suspended in a central position of their quarter, about 1 m from the observation chamber and about 5 cm above the bottom, using ropes attached to the roof construction and the tank walls.

For surveillance of the acoustic field, a hydrophone was positioned in the centre of every quarter. During experiments sound measurements were carried out with the hydrophones in quarter 1 and quarter 4, while the hydrophones in quarter 2 and 3 were only used as dummies. A fifth moveable hydrophone was used while measuring the acoustic field in the tank.

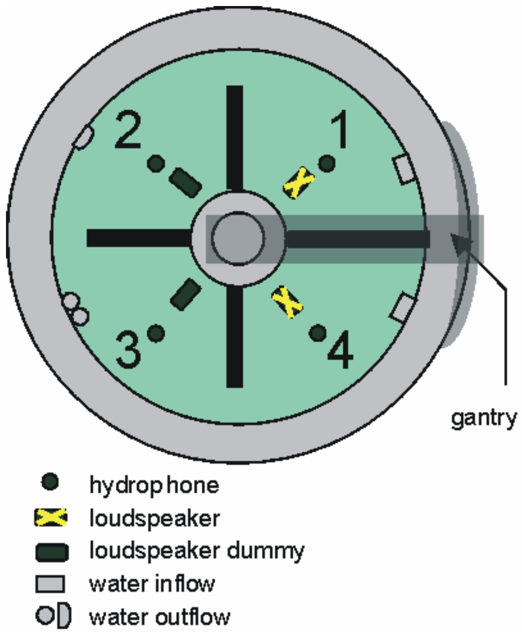


Fig. 11: Sketch of the tank equipment. The experimental tank is divided into quarters by one long barrier extending from the inner to the outer wall and three shorter barriers that let the fish move between the quarters at the periphery. The quarters in the sketch are marked with numbers. Quarter 1 and 4 are equipped with loudspeakers, quarter 2 and 3 with loudspeaker dummies. Every quarter contains a hydrophone. The gantry leading to the observation chamber rests above the long barrier between quarter 1 and 4. The water inflows are in quarters 1 and 4, the water outflows in quarters 2 and 3.

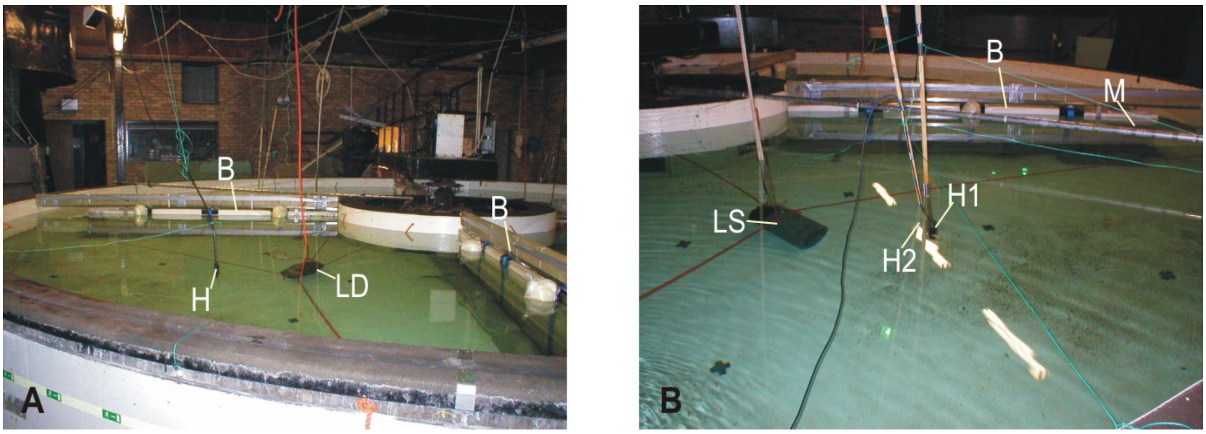


Fig. 12: **A**: Tank quarter 2. A loudspeaker dummy (LD) and a hydrophone (H) can be seen in the centre of the quarter. The quarter is limited by barriers (B). **B**: Tank quarter 1. The loudspeaker (LS) can be seen on the left hand side. The stationary hydrophone 1 (H1) is in the centre of quarter 1 next to the moveable hydrophone (H2) that was used for measurements comparing hydrophones. In front of the barrier (B) the metal bar (M) is visible to which the mobile hydrophone 6050C was attached while measuring the acoustic field in the tank.

3.3.1 Sound barriers

The four barriers served to amplify the sound pressure difference in the tank. In small tanks (compared to the wave length of the sound) sound attenuation is very small (see chapter 1.2.3.3) especially at low frequencies making sound insulation nearly impossible.

The sound barriers were constructed from marine plywood boards to which Styrofoam plates were attached on both sides (Fig. 13A), plastic sheeting were wrapped around for protection (Fig. 13B). In order to prevent warping, belts tightened by ratchet buckles were used (Fig. 14). The ratchet buckles were wrapped in plastic bags filled with foam to avoid injuries to the fish as well as additional sound reflection and corrosion. Due to the high buoyancy of the Styrofoam barriers, each one was kept in place using a steel bar and wooden beam positioned above the barrier, and steel brackets weighted with gravel-filled bags at the base (Fig. 15).

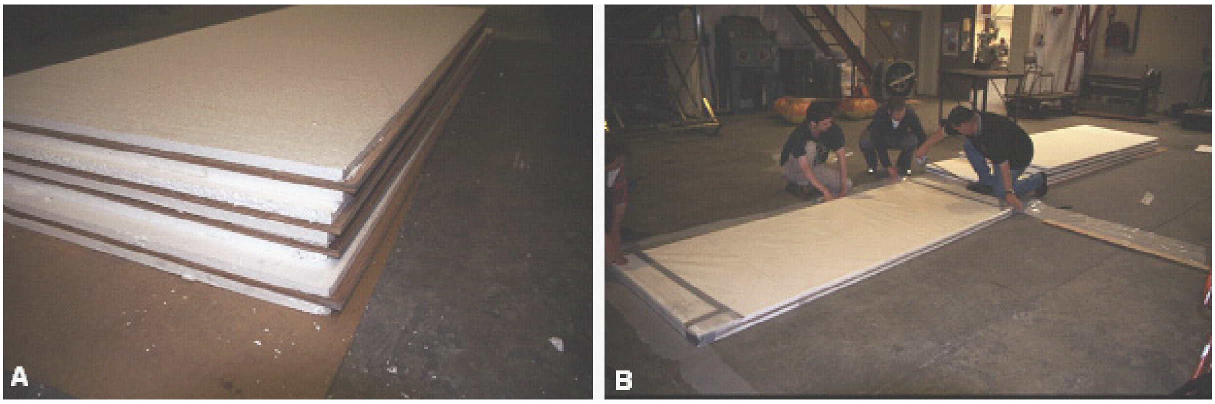


Fig. 13: Barrier construction. **A**: Structure of the barriers. The four barriers consisting of a marine ply board and two Styrofoam sheets lie one upon another. **B**: The barriers are wrapped in plastic sheeting and sealed with tape.

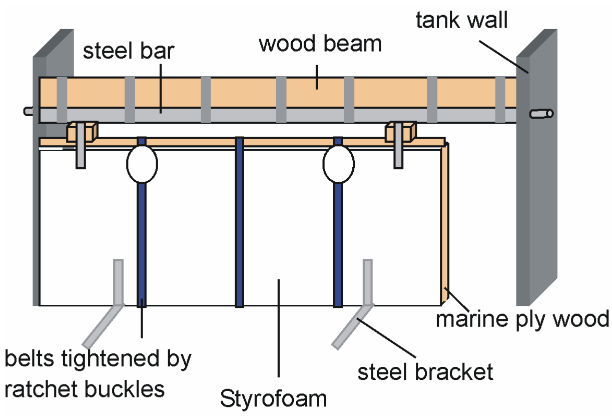


Fig. 14: Sketch of the barrier construction in the tank. The barriers are hold in place by a steel bar anchored in the upper part of the tank walls and steel bracket at the bottom of the tank. The steel bar is stabilized against bending by a wood beam. Belts tightened by ratchet buckles are used to take steps against the buoyancy of the Styrofoam.

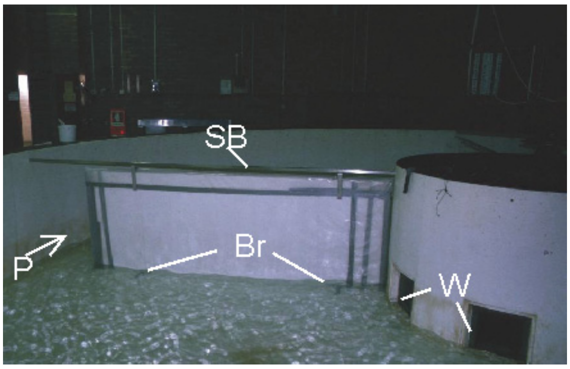


Fig. 15: Anchorage of a barrier. The tank was nearly emptied to anchorage the barriers in the upper part of the tank wall by a steel bar (SB). In the lower part of the barrier steel brackets (Br) keep the barrier in position. On the left hand side the passage (P) is visible where the fish can pass through to move between the different quarters. On the right hand side two windows (W) in the observation chamber can be seen.

Styrofoam was used for its sound isolating effect. There is minimal sound transport between water and air due to the different impedance of the agents. Due to its high air content Styrofoam is very efficient at reflecting sound.

3.3.2 Sound production and measurement

For sound production, two Type J-11-Audio-Frequency Transducers (*USRL US-Navy*) were used. The J-11 is a monopole sound source and emits sound evenly in all directions at low frequencies. The loudspeakers were connected to a 25 WT amplifier (*Derritron*) to adjust the sound level. An Sweep/Function Generator, Model 506 (*Exact*) was used to adjust the frequencies.

In general loudspeakers, such as the J-11 are only used under water for a short period of time. To prepare the loudspeakers for the longer term operation needed, parts vulnerable to corrosion were replaced.

Prior to the experiments, the sound production of loudspeaker 1 was tested at different frequencies and voltages. This test showed a decrease of sound level below 100 Hz. Apart from this the sound production was relatively even at all voltages (Fig. 16). The frequency 25 Hz could not be produced at a sound level of 140 dB re 1µPa.

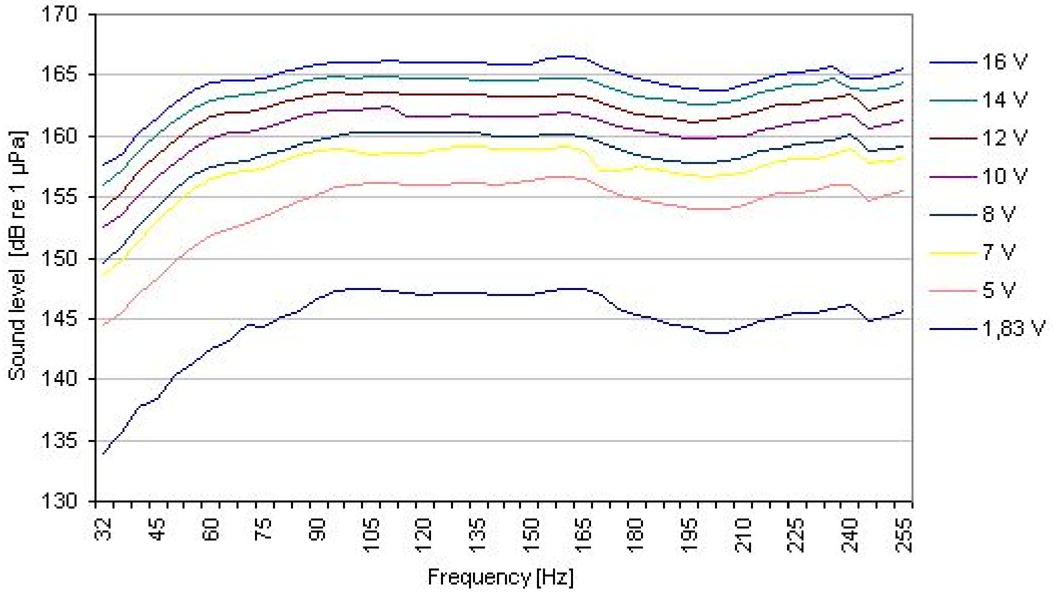


Fig. 16: Sound levels produced by the J-11-loudspeaker LS 1 at different frequencies and voltages.

Three hydrophones were used to measure the sound fields in the tank. Two hydrophones were stationary in quarter 1 and quarter 4 as reference hydrophones next to the loud-speakers; hydrophone 1 (H1) a TC4013 (*Reson*) at a distance of 0.72 m from the centre of loudspeaker 1, and hydrophone 4 (H4) a 6050C (*ITC*) at 0.96 cm from the centre of loudspeaker 2. Hydrophone 5 (H5), a 6050C (*ITC*) could be moved around to determine the acoustic field in different positions of the tank.

A VP1000 Voltage preamplifier (Reson) and a Low Noise Amplifier Type 450 (*Brookdeal-Electronics*) amplified the input from TC4013. SR 560 amplifiers (*Stanford Research Systems*) were used to amplify the ITC-6050C signals. The TC4013 was connected to a LQ100/30 (*Countant*) for energy supply whilst the 6050Cs were supplied with energy from a Stanford amplifier. An oscilloscope HP 54501A (*Hewlett Packard*) was used to read the frequencies and the voltage as rms (root mean square) to calculate the sound levels. The waveforms of both hydrophones could be controlled simultaneously on the display of the oscilloscope.

Two computer programmes were used for the evaluation of the sound measurements. With the programme "R" 1.8.0 every single reading is displayed. Different colours depending on the sound level make it possible to get an impression of the sound field in the tank. Extra-ordinary readings attract attention by different colouring. With „MATLAB" 6.1 a surface is calculated from the readings by interpolation. The surface is smoother and gives a good impression of the overall sound field in the tank. However extraordinary readings are smoothened as well and can easily be missed.

3.3.3 Selection of produced sounds

The highest sound levels are emitted by offshore wind turbines at a frequency range between about 25 and 250 Hz. Higher frequencies emitted by the wind turbines are masked by background sound produced by wind, waves, current and other sound sources (DEGN 2000). Measurements on 2-MW offshore wind turbines showed sound levels of 130 dB re 1µPa at a frequency of 25 Hz. Future offshore wind turbines are planned of up to 6-MW which makes it likely that higher sound levels will be generated.

On the basis of these data acoustic experiments were carried out at five different low frequencies of 25 Hz, 60 Hz, 90 Hz, 125 Hz and 250 Hz at two sound levels of 130 dB re 1µPa and 140 dB re 1µPa (Table 3).

The used frequency range is included in the hearing range of cod and plaice (BUERKLE 1967, 1968, CHAPMAN & HAWKINS 1973, CHAPMAN & SAND 1974, OFFUT 1974, KARLSEN 1992a). It also includes communication frequencies of the cod (HAWKINS & RASMUSSEN 1978).

Table 3: Frequencies and sound levels used in the experiments and the reasons for their use

	Criterion for choice
Frequency	
25 Hz	Frequency with high sound levels from 2-MW offshore wind turbines (DEGN 2000, Fig. A 1 and A 2, appendix).
60 Hz	Lower end of the most sensitive hearing range of cod (SAND & HAWKINS 1973).
90 Hz	The grunts of cod used in different social contexts have their greatest energy at about 95 Hz and its harmonics (HAWKINS & RASMUSSEN 1978). Frequency with rather lower sound levels measured at offshore wind turbines.
125 Hz	Frequency emitted in higher sound levels by 450-KW offshore wind turbines (DEGN 2000).
250 Hz	Top end of the hearing range of plaice (CHAPMAN & SAND 1974).
Sound pressure level [dBre 1 µPa]	
130 dB	Calculated sound levels of 1.5-MW offshore wind turbine at 1 m distance from measurements at a distance of 83 m from the turbine (INGEMANSSON 2003, Fig. A 3, appendix). Predicted sound level of 2-MW offshore wind turbine (DEGN 2000).
140 dB	Sound level expected from 4.5-MW offshore wind turbines by modelling (DEWI 2004, Fig. 1).

The wavelengths of the frequencies used are between 6 and 58 m (Table 4), which clearly exceeds the annular radius.

Table 4: Sound wave lengths of the tested frequencies

Frequency	Wavelength
25 Hz	57.60 m
60 Hz	24.00 m
90 Hz	16.00 m
125 Hz	11.52 m
250 Hz	5.76 m

3.3.4 Initial sound measurements

The reference for the unit “dB” used for sound levels in this report is 1 µPa as usually used in underwater sound measurements unless another reference is given. The sound levels produced in the tank were defined as the sound level measured at the reference hydrophone next to the used loudspeaker. Therefore using the term “at a sound level of 130 dB re 1µPa” in following means the sound field existing in the tank while a sound level of 130 dB re 1µPa was measured at the reference hydrophone.

Initial comparison measurements were made between the TC4013 (H1) and the 6050C (H5) by suspending them in the same place of the tank (Fig. 12B). The arrows in Fig. 17 show the direction of the tip of H5 in different measurements.

The two hydrophones show differences of about 1.5 – 4 dB re 1µPa depending on the frequency (Fig. 17). The readings of H5 exceeded those of H1. The differences were not related to the orientation of H5, but to the frequency. The difference between the two hydrophones was considered small enough compared to the sound pressure difference in the tank to regard them as equally sensitive.

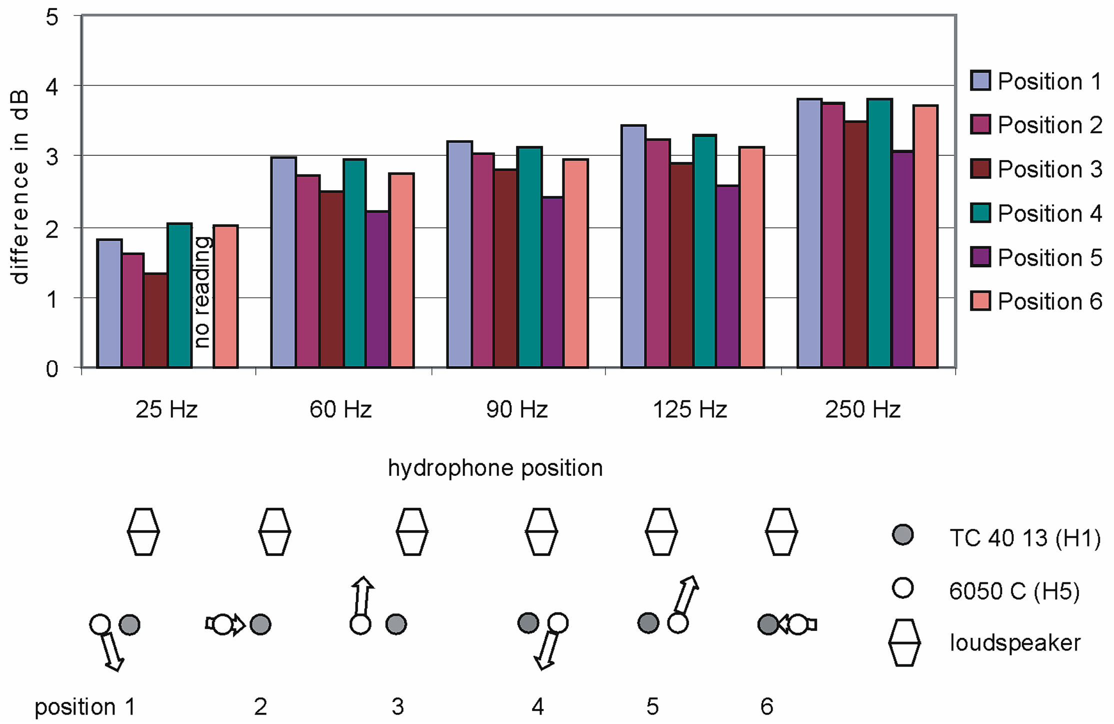


Fig. 17: Difference of the readings of H1 (TC4013) and H5 (6050C) at various frequencies and orientations of H5. The different orientations of H5 were caused by a diagonal position of the hydrophone in the water column. The arrows show the direction of the tip of the hydrophone during measurement. Differences increased with frequency but were always below 4 dB.

3.3.5 Hydrodynamic measurements

Attempts were made to measure the hydrodynamic field in the tank. Unfortunately the method proved to be unsuccessful and the results did not provide a description of the hydrodynamic field in the tank. Nevertheless, a description is given of what has been done.

The particle displacement in three dimensions was measured using a geophone PE-6/B (*Sensor Nederland*) containing three oscillation sensors (Fig. 18). Due to the position of the sensors readings can be taken simultaneously in three orthogonal axes.

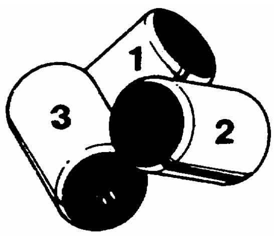


Fig. 18: Sensor positions in the geophone PE-6/B (from the geophone data sheet).

The data from the sensors were recorded by an PR6-24 Portable Field Recorder (*Earth Data*). The data rate was 750 reading per second from each sensor, stored as a new data file every 15 minutes. Since the readings were taken continuously it was necessary to take the exact time when the measurements of certain frequencies and sound levels took place to relate the readings to the sound field produced in later evaluations. Automatic synchronization was not possible and the exact time was read from the Internet and the time difference between real time and internal clock of the geophone was taken into account in data evaluations.

To let the geophone float in the water column with neutral buoyancy, it was attached to a polyethylene tube (23 cm diameter 43 cm in length) by a metal clamps to equalize the weight of the geophone (590 g) (Fig. 19A). Six small screws at the top of the tube and two large screws at the bottom were added to achieve neutral buoyancy with the geophone floating 1-5 cm above the tank bottom.

A string was attached to three points at the top of the polyethylene tube. The cable of the geophone was fed through the top of the tube and fixed to a slit in the lower part of the tube to avoid interference with the geophone movement that could lead to water leaking into the geophone.

Above the water surface, a metal bar resting on the inner and outer tank wall and insulated against vibration by rubber foam was used to determine the position of the geophone using the attached string. While the geophone was floating above the tank bottom (Fig. 19B) the current next to the water inflow caused shifting movements that needed to be reduced by a screw used as a weight attached to a 1 m string at the bottom of the floating device. To determine the direction of the sensors, the position of the cable indicating the north-south direction was registered at each measurement point.

19 measurement points were determined in quarter 4 (Fig. 20). The measurements were carried out using loudspeaker 2 in quarter 4 because this loudspeaker was used in all but one of the behavioural experiments.

Eleven readings (five frequencies, two sound levels one background noise level) were carried out at every point. Every reading was taken for a duration of 30 seconds. For analysis, the 15 second periods starting from the 11th second were taken. These 15 seconds contained 11250 data points each.

The results of the measurements were evaluated with the programmes DaDisp 2002 and DaDisp_SE 2002. The 15 minutes files produced for every sensor were broken down into smaller files matching the different sound fields produced for evaluation. The programme calculated the RMS as a measure for the amplitude of the oscillation. The RMS was displayed in digits and the voltage was calculated from 1 digit equalling 0.1 µV. Using the

RMS voltage the formula $\xi = \frac{RMS}{2 \cdot \pi \cdot f \cdot 29}$ was used to calculate the particle displacement including the frequency and a transfer constant at 1 Hz related to the sensors.

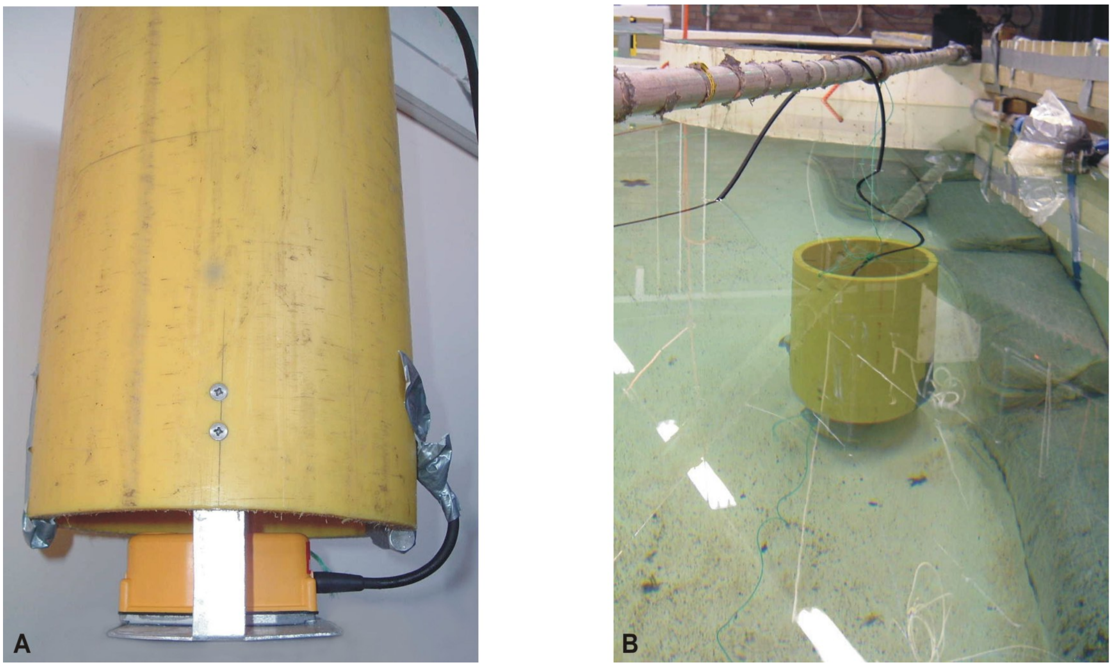


Fig. 19: **A**: Geophone attached to a polyethylene floating device **B**: Geophone floating above the tank bottom.

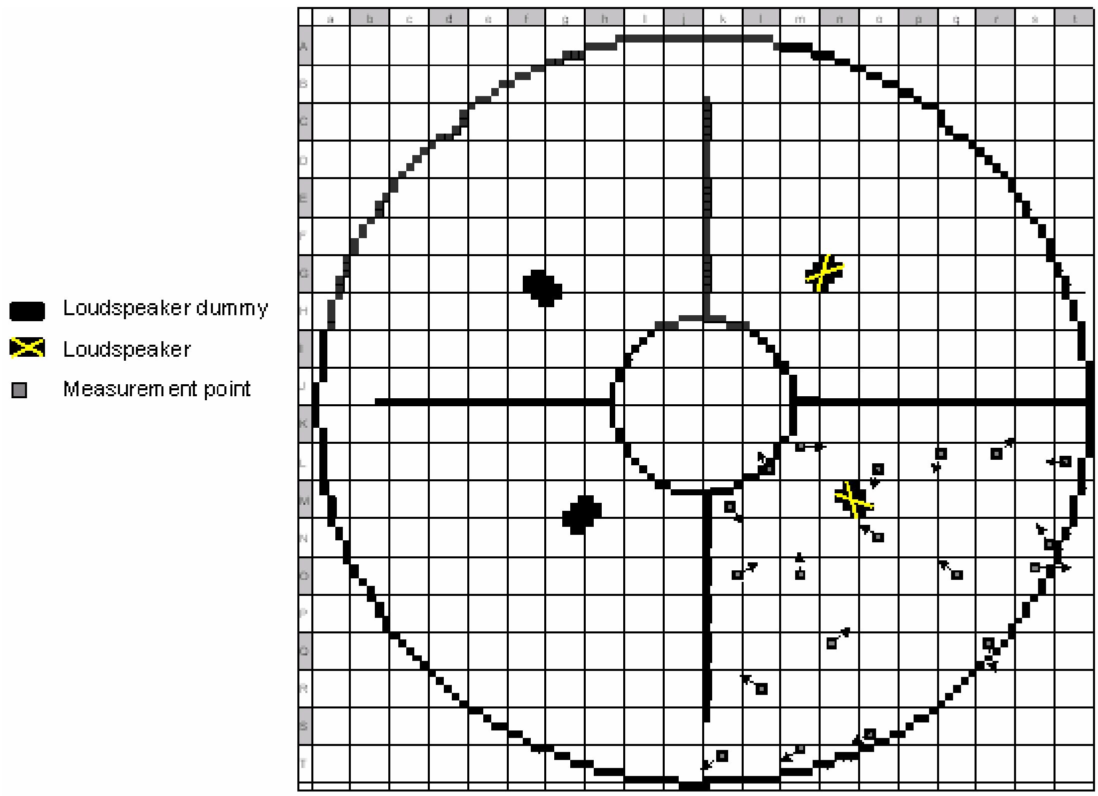


Fig. 20: Sketch of the tank with positions of 19 geophone measurement points to determine the particle displacement. The sound was produced by loudspeaker 2 in quarter 4. The arrows show the direction of the cable indicating the north-south direction of the geophone.

It turned out that the data collected were not suitable for evaluation and since the geophone was not available for further measurement results cannot be presented.

3.4 Video surveillance equipment

To study the behaviour of the fish, four cameras were installed about 5 m above the tank capturing one frame per second in an AWLive-file to a computer hard drive, using the video surveillance programme “Active Web Cam 5.0”. Each Digital *Panasonic* WV-CL350 camera (2.8 mm lens) was positioned so as to focus on one quarter of the tank.

3.5 Experimental set up

3.5.1 Sound experiments

Experiments were carried out using juvenile and adult plaice and cod. Each group of fish was subjected to an experimental series of nine to ten experiments exposing them to differing frequencies and sound levels. For each experiment the fish were exposed to the sound continuously for 24 hours. In between the experiments, five days were allowed for recovery. The order of test frequencies and intensities was chosen by random (Table 5).

Table 5: Order of experiments carried out with juvenile and adult cod and plaice

No. of experiment	Frequency [Hz] and sound level [dB re 1µPa] of the experiments		Frequency [Hz] and sound level [dB re 1µPa] of the experiments	
	juvenile cod	adult cod	juvenile plaice	adult plaice
1	90 Hz 140 dB	90 Hz 130 dB	60 Hz 140 dB	25 Hz 130 dB
2	60 Hz 130 dB	125 Hz 140 dB	25 Hz 130 dB	60 Hz 130 dB
3	125 Hz 140 dB	250 Hz 140 dB	125 Hz 130 dB	60 Hz 140 dB
4	125 Hz 130 dB	25 Hz 130 dB	250 Hz 140 dB	90 Hz 130 dB
5	25 Hz 130 dB	60 Hz 140 dB	60 Hz 130 dB	250 Hz 140 dB
6	90 Hz 130 dB	90 Hz 140 dB	90 Hz 130 dB	90 Hz 140 dB
7	25 Hz 140 dB	25 Hz 140 dB	250 Hz 130 dB	125 Hz 130 dB
8	250 Hz 140 dB	60 Hz 130 dB	125 Hz 140 dB	125 Hz 140 dB
9	60 Hz 140 dB	125 Hz 130 dB	90 Hz 140 dB	250 Hz 130 dB
10				25 Hz 140 dB

3.5.2 Habituation in plaice

To investigate the effect of sound on the settlement behaviour of plaice during the acclimatization period, an additional experiment was carried out on six juvenile plaice (TL 24 to 30 cm) released into the tank while sound at a frequency of 60 Hz at a sound level of 140 dB re 1µPa was produced in quarter 4. One fish was released into quarter 1, one into quarter 4 and two fishes each into quarter 2 and quarter 3. The sound was switched off after 45 hours. The distribution of fish was evaluated for a further 14 hours. The results of this experiment were compared with the acclimatization periods of the experiments with juvenile and adult plaice.

3.6 Evaluation of videos

3.6.1 Distribution and behaviour

Video was recorded continuously at a rate of one frame per second over the whole experimental period of about 9-10 weeks each. The 24 hour sound period and the 24 hours periods before and after sound exposure were examined for changes in behaviour and distribution caused by the sound.

The behaviour and the distribution of fish in the tank were analysed manually every 15 minutes. Small marks on the bottom of the tank divided each quarter in twelve sections (Fig. 21), which made it possible to determine the distribution of fish in the quarters. The fish showed a general preference for tank quarter 4. Therefore the evaluation was mostly restricted to this quarter. In addition to the distribution in quarter 4, the behaviour was recorded as “resting” and “active”. “Active” was defined as directed swimming movement as opposed to small movements during resting behaviour and turning on the spot. It was divided into swimming at the bottom, midwater or close to the surface but this distinction was not used in the further evaluation.

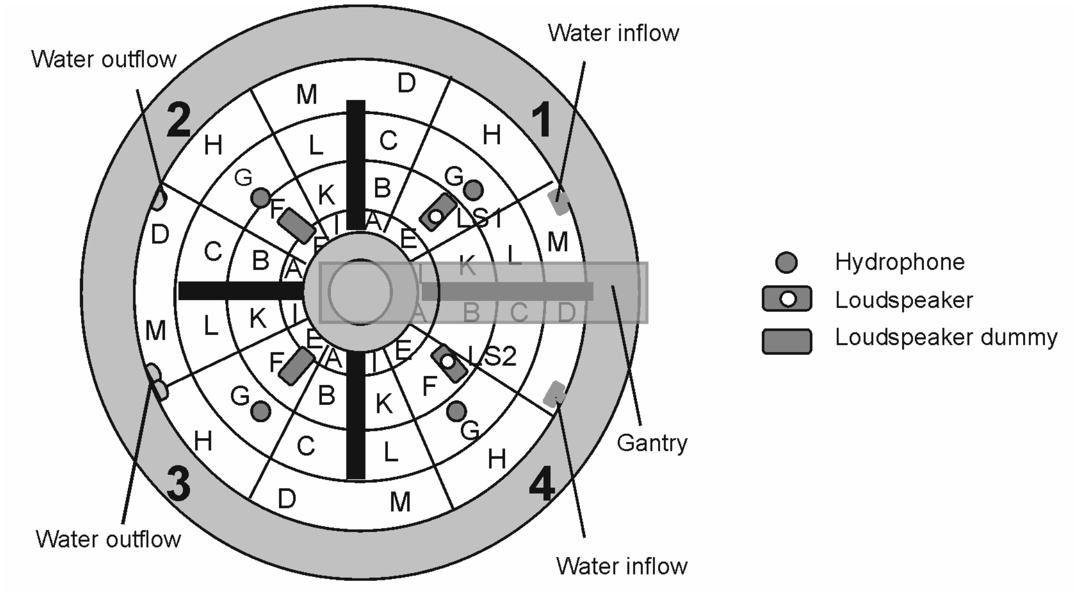


Fig. 21: Sketch of the tank showing the sections to determine the position of fish.

For display of the video results the programs "SigmaPlot 2001", “Corel Draw 10” and "MS Excel 2003" were used. Statistical analysis was carried out using “OpenStat 4”.

The percentage of fish in the quarter where the sound was produced was determined every 15 minutes. The data are displayed for 72 hours including three 24 hours periods before, during and after sound production. The raw data collected in every 72 hours period are displayed in four different figures for every experiment (Fig. A 26 to Fig. A 63, appendix). Two figures (a + c) show the percentage of fish counted every 15 minutes overall and divided into “resting” and “active” . The same data are displayed as box whisker plots (b + d) with data separated into day- and night-periods and “resting” and “active”. Additional box plots displaying the total fish numbers in the periods before, during and after sound production can be found in the result chapters 4.2.1.2 and 4.2.2.2. The box displayed in the box-whisker-plots contains 50% of the data, the line in the box marks the median. Additionally marked by whiskers are the 10. and 90. percentile and the 5. and 95. percentile marked by asterisks.

In an initial analysis all experiments for each fish group were considered to be replicates and the Friedman-Test (α = 5%) was used to check for an overall effect of sound. For this test, average fish numbers differing by less than 2% between the three time periods were considered to be equal and therefore given equal ranks. Following this, the Kruskal-Wallis-H-Test (α = 5%) was used to check for differences between the three 24-h-periods in each experiment. If the number of fish in the experimental periods differed significantly the Mann-Whitney-U-Test (α = 5%) with Bonferroni correction was used to compare each period with the two others. The results of the Kruskal-Wallis- and Mann-Whitney-test are given in the Table A 6 and Table A 8 in the appendix. Differences between day and night in 24 hour periods were also tested for significance using the Mann-Whitney-U-Test (α = 5%) (Table A 5 and Table A 7, appendix). To compensate for the low activity in plaice only one observation per hour was entered into the statistical evaluation.

3.6.2 Loudspeaker vicinity analysis

The sound level decreased very quickly with distance from the sound source. For this reason an additional evaluation of the immediate vicinity of the sound source with sound levels attaining 124 to 161 dB re 1µPa was carried out in some experiments to examine whether these high sound levels might cause reactions that do not appear in more distant parts of quarter 4.

A circle of one meter around the loudspeaker was drawn to count every fish entering this circle in a given period of time (Fig. 22). Additionally the duration of stay in this circle was measured for every visit. This evaluation was made before, during and after sound production in the initial and final 30 minutes of every 24 hours period. It was carried out for three experiments with plaice and four experiments with cod. The fish numbers observed in the circle was higher than the fish numbers in the tank due to numerous revisits of fish in the area.

Fig. 22: Tank quarter 4: Circle marks the area around loudspeaker 2 that was used for the loudspeaker vicinity evaluation.

4 Results

4.1 Acoustic field in the experimental tank

4.1.1 Acoustic field with sound barriers

The acoustic field in the experimental tank divided by barriers, was measured at 86 points (Fig. 23). At each point sound levels were measured at four depths (0 m; 0.4 m; 0.8 m; 1.2 m from the bottom of the tank).

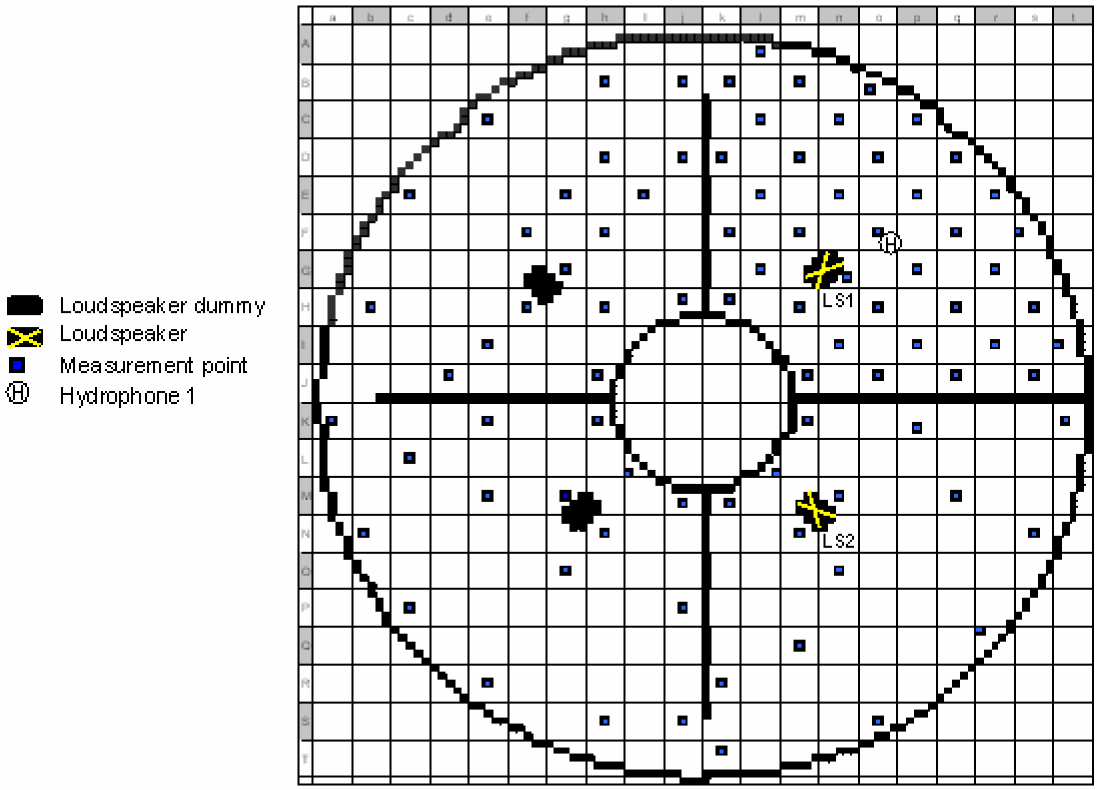


Fig. 23: Position of measurement points in the experimental tank divided into quarters by sound barriers. The sound was produced by loudspeaker 1 in quarter 1. The measurements were made at 86 measurement points at four different water depths with a 6050C hydrophone.

In quarter 1, where the sound was produced in this analysis, measurements were made at 37 points. The sound level decreased very quickly around the sound source while the differences in sound levels between the measurement points in the quarters 2-4 were relatively small. Therefore the measurements in the quarters 2-4 could be reduced to 15 to 18 points. In accordance with the preference of many fish for more protected areas the majority of the measurement points were located in the vicinity of the tank walls, the loudspeakers and loudspeaker dummies.

Each of the 86 measurement points contained 40 readings (measurements of different sound fields at four water depths). Unusual readings were checked with additional measurements afterwards. The readings of hydrophone 1 (H1) at a distance of 0.72 m from loudspeaker 1 were used as reference for the produced sound level.

With few exceptions, the frequency produced by loudspeaker 1 could be determined at all points in the tank. But especially at low frequencies the purity of the frequency decreased with the distance from the sound source.

As an example for the acoustic field in the tank Fig. 24 and Fig. 25 show the results 0.4 m above the tank bottom during production of 25 Hz at a sound level of 130 dB re 1µPa. The measurements are displayed as the difference from the sound level read at hydrophone H1 (Fig. 24) or as absolute sound levels (Fig. 25).

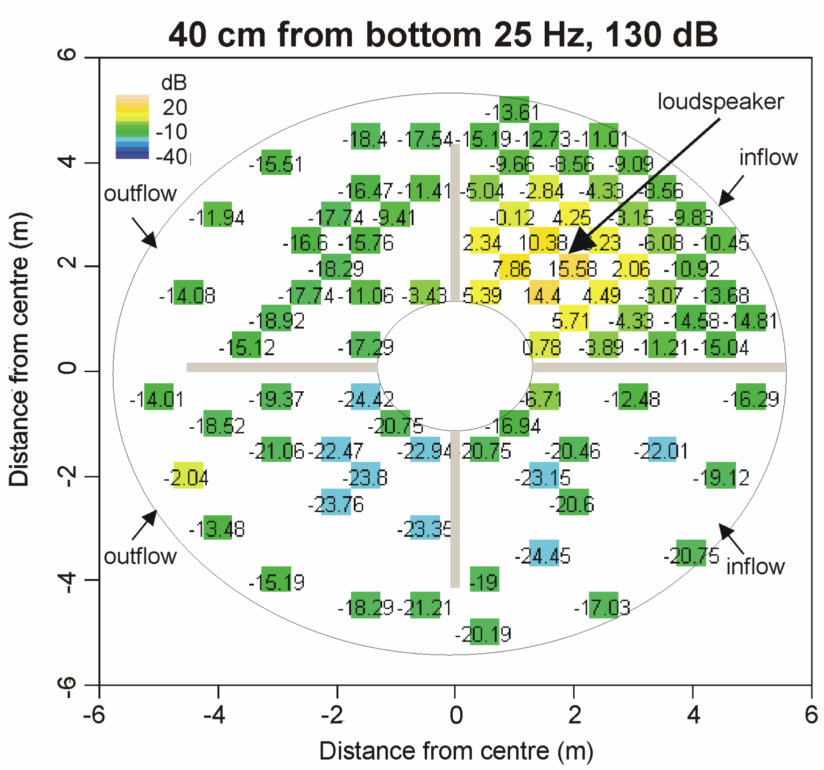


Fig. 24: Sound field in the experimental tank 0.4 m above the bottom of the tank during sound production of 25 Hz at a sound level of 130 dB re 1µPa displayed with “R”. The measurements are displayed as the difference from the reference hydrophone H1, which was located at 0.72 m distance from the sound source. The sound was produced with loudspeaker 1 in quarter 1 (above right). In the vicinity of the loudspeaker sound pressure exceeded the reference sound level. With distance from the loudspeaker the sound decreased rapidly. In the other quarters the sound levels were much lower, sound levels being lowest in quarters 3 and 4 (bottom left and right). Close to the outflow in quarter 3 (bottom left) the sound level increased by more than ten decibel.

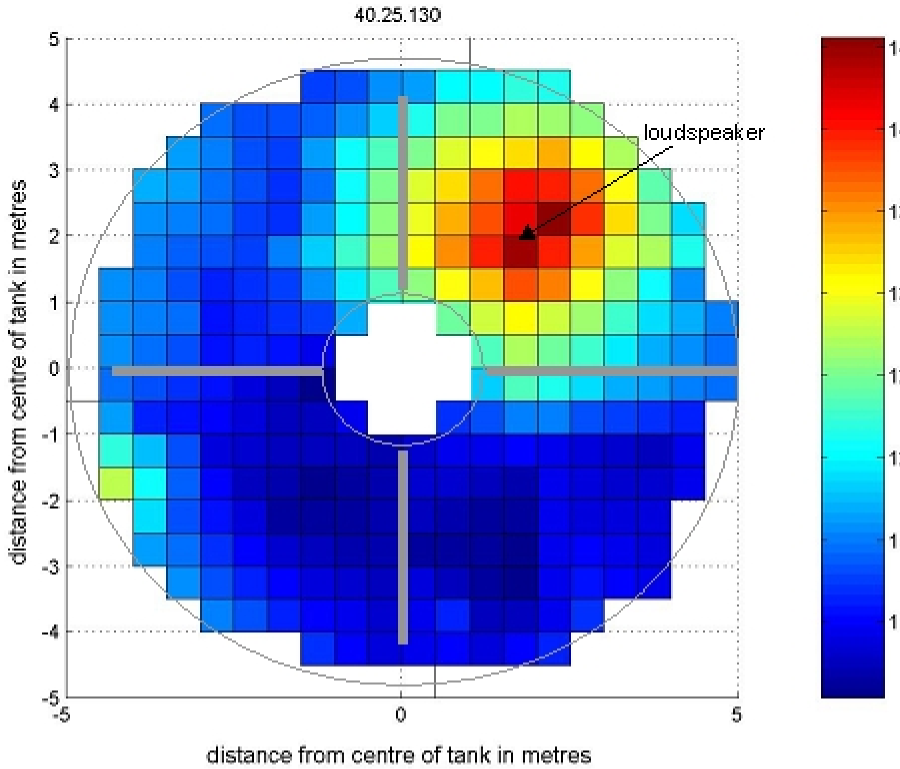


Fig. 25: Same as Fig. 24 but absolute sound levels displayed with “MATLAB”. The interpolation of nearby measurements caused the visual obliteration of an effect of the sound barriers.

The sound decreased rapidly with distance from the sound source in quarter 1. Differences of about 25 dB appeared between the reading next to the loudspeaker and readings next to the outer tank wall. Close to the sound barriers the decrease could come to about 30 dB. In quarter 2 the sound level decreased further with an average decrease of -14.82 dB compared with the produced sound level. Highest sound levels could be observed close to the sound barrier to quarter 1 and along the outer tank wall. In quarter 3 a further decrease of sound level to -19.04 dB on average could be observed with lowest sound levels in the middle of the quarter and next to the inner wall. The water outflow caused a regional sound peak. In quarter 4 the sound level decrease came to –18.66 dB on average with lowest values in the centre of the quarter. The lower sound decrease in quarter 2 could be explained by the direct connection between quarter 2 and quarter 1 where the sound was produced. Quarter 4 was separated from the sound source by the long barrier, which reduced sound transport into quarter 4 and caused lower sound levels. Quarter 3 had the greatest distance to the sound source and lowest sound levels were expected.

One measure for the sound field in the tank was the sound pressure difference calculated by the difference between the minimal and maximal values. For 25 Hz at a sound level of 130 dB re 1µPa it was 40 dB at a water depth 0.4 m above the tank bottom. At other frequencies and sound levels in different water depths the pressure difference reached between about 32 dB and 52 dB. A compilation of the minimal and maximal readings at different frequencies and sound levels and the calculated sound pressure differences can be found in Table A 2 (appendix), detailed results of the acoustic field measurements are shown in Fig. A 4 to Fig. A 25 (appendix).

Sound levels decreased from the bottom of the tank to the water surface. The differences between bottom and surface generally came to 12 to 15 dB. This result applied to the produced sound fields as well as to the background noise. Looking at the different frequencies the sound pressure difference close to the bottom of the tank reached between 41 and 51 dB whereas close to the surface 32 and 44 dB were attained in accordance with the lower sound levels measured near the surface.

Sound level decrease towards the Styrofoam and concrete wall was compared since the acoustic properties of concrete and Styrofoam are different from each other with concrete being a rigid and Styrofoam being a soft boundary towards water. From the centre of the tank towards the Styrofoam wall the sound level decreased in all sound fields. Towards the outer concrete wall the sound level decreased (Fig. 26) except for the two most external points (10 cm and 60 cm from the tank wall) at 250 Hz where a difference became visible as a lack of further sound level decrease close to the concrete wall. At 90 Hz the sound level decrease in direction of the concrete wall was clearly smaller than the decrease in direction of the Styrofoam barrier.

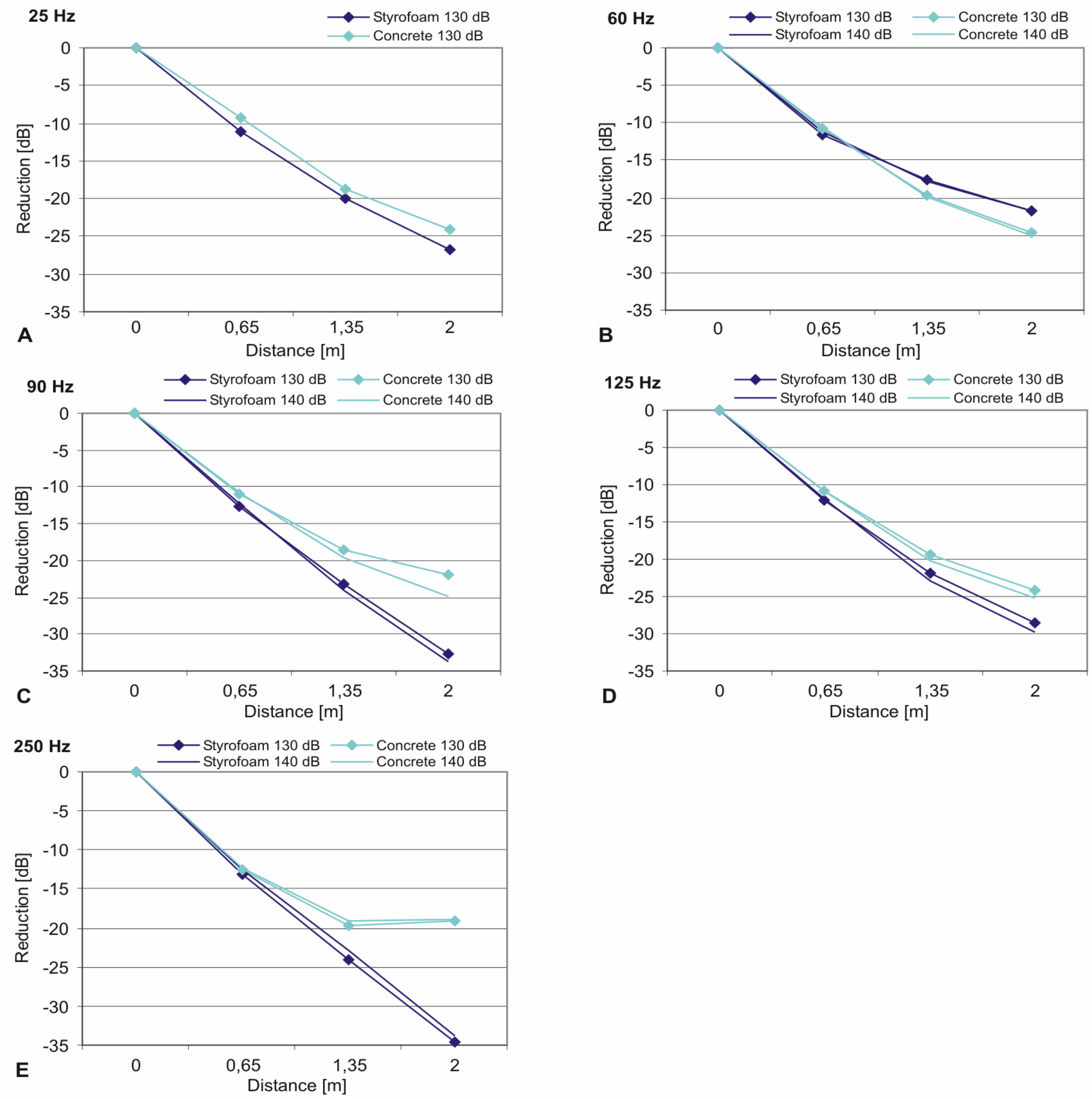


Fig. 26: Sound level decrease from the sound source towards the concrete wall and the Styrofoam barrier in different frequencies and sound levels, measured as a function of the distance from the sound source. For the frequencies 25, 60 and 125 Hz (**A, B, D**) the differences in sound level decrease were small with slightly stronger decrease closer to the sound source. At 250 Hz (**E**) sound levels showed no decrease close to the concrete wall. At 90 Hz (**C**) smaller sound level decreases were found in the direction to the concrete wall than to the Styrofoam barrier.

Beyond slightly higher sound levels in the vicinity of the outflows a remarkable deviation from the general strong decrease in sound level with distance from the loudspeaker were comparatively higher levels in quarter 3 close to the observation chamber at frequencies of 90 Hz and above (Fig. 27). Since measurement errors were ruled out by additional measurements it is likely that the higher sound levels were generated by sound waves travelling trough the concrete wall of the observation tube.

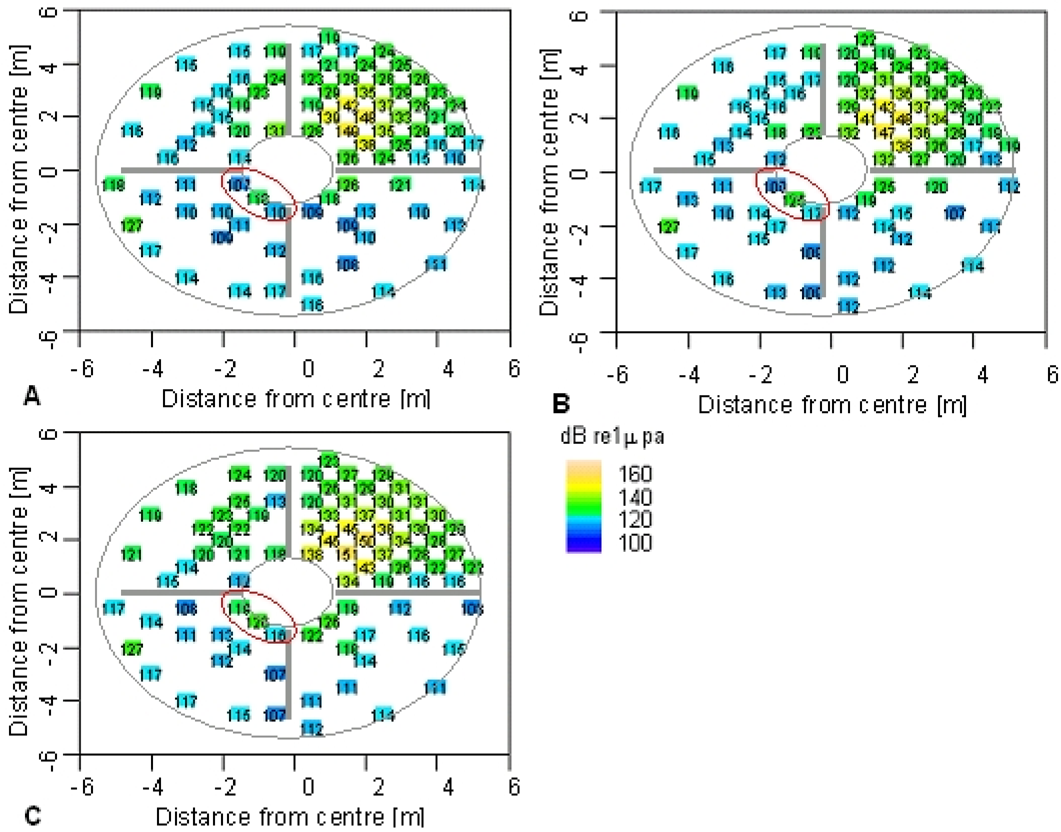


Fig. 27: Sound field at 0.4 m above the tank bottom displayed as absolute readings during sound production of **A**: 90, **B**: 125 and **C**: 250 Hz at a sound level of 130 dB re 1µPa at 0.72 cm distance from the sound source. Extraordinary high readings in quarter 3 next to the observation tube are marked red.

4.1.1.1 Sound field using a sound source in quarter 4

While the tank was equipped with a second sound source (LS 2) located in quarter 4 additional sound measurements in quarter 4 were carried out using LS 2. The sound field produced in quarter 4 by loudspeaker 2 resembled the results in quarter 1 while using loudspeaker 1 (LS 1) as sound source (Fig. 28). For this reason measurements could be limited to 17 measurement points in quarter 4 containing the sound source. Higher readings in quarter 4 compared with the readings in quarter 1 using LS 1 could be explained by a slightly greater distance between sound source and reference hydrophone. In quarter 4 it was 0.96 m, while it was 0.72 m between loudspeaker 1 and the reference hydrophone 1 in quarter 1. Due to the higher distance a slightly higher sound production at loudspeaker 2 was necessary to get the reference intensity at the hydrophone.

The position of loudspeaker 2 was about 0.2 m closer to the long Styrofoam barrier than it was in quarter 1. This caused higher sound levels along the long barrier in quarter 4. Fig. 28 shows the readings during sound production of 25 Hz and 130 dB re 1µPa as an example for the sound field in quarter 4. More figures showing the acoustic field during sound production in quarter 4 can be found in Fig. A 13 to Fig. A 17 (appendix).

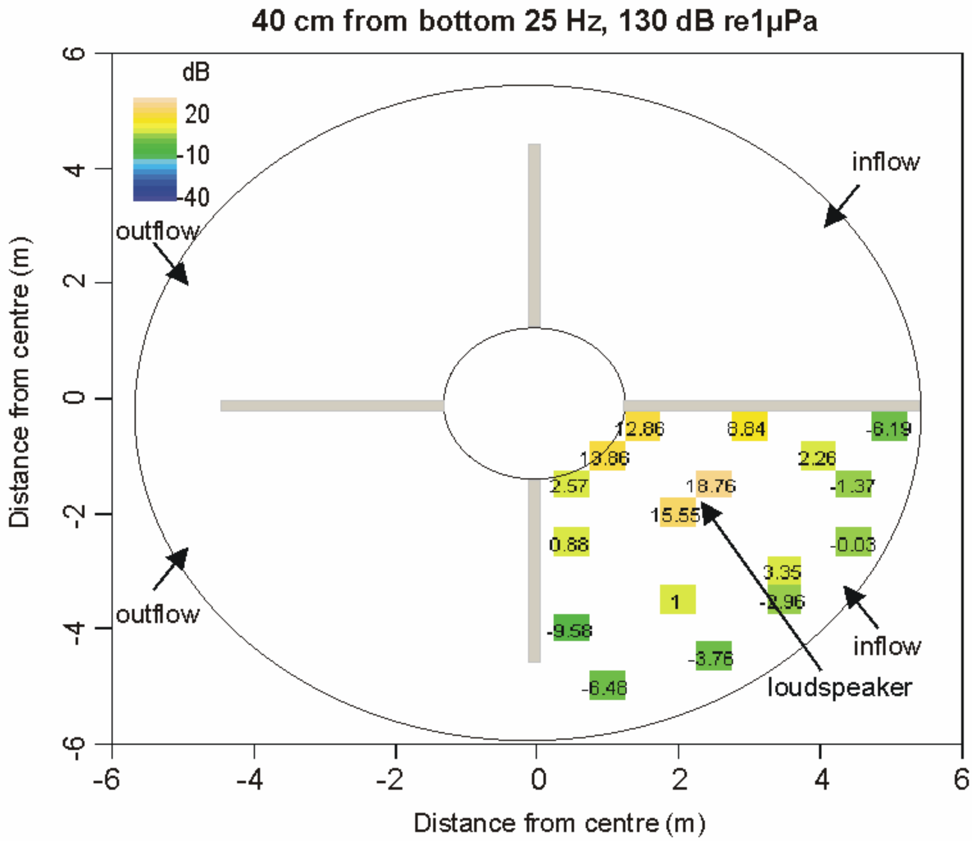


Fig. 28: Sound field in quarter 4 at 17 measurement points 0.4 m above the bottom of the tank during sound production of 25 Hz at 130 dB re 1µPa. The sound was produced with loudspeaker 2 in quarter 4 (bottom right). The numbers indicate the difference from the sound level measured at hydrophone 4 at 0.96 m distance from the sound source. A rapid sound level decrease can be seen around the loudspeaker. The measurements in the vicinity of the long barrier are higher than those close to the barrier in direction to quarter 3 due to the position of the loudspeaker closer to the long barrier.

4.1.1.2 Background noise

Sound levels were also measured in the tank without sound production to compare the background sound permanently existing in the tank (Fig. 29A) with the produced sound fields. Sound levels of on average 99 dB re 1µPa at the surface to 112 dB re 1µPa at the bottom of the tank were measured with sound level variations evenly distributed throughout the tank with slightly higher sound levels in the vicinity of the tank walls and lower levels in the centre of the quarters. In comparison the average sound levels in quarter 1 during sound production of 25 Hz at 130 dB re 1µPa were 112 dB re 1µPa at the surface and 128 dB re 1µPa at the bottom. To illustrate the difference between background sound and sound production Fig. 29B shows the absolute sound levels during production of 25 Hz at 130 dB re 1µPa. The difference is most obvious in quarter 1 with high sound levels in most areas of the quarter decreasing with distance from the sound source. The background sound measurements only show local sound pressure peaks and the sound field in quarter 1 is comparable with the other parts of the tank

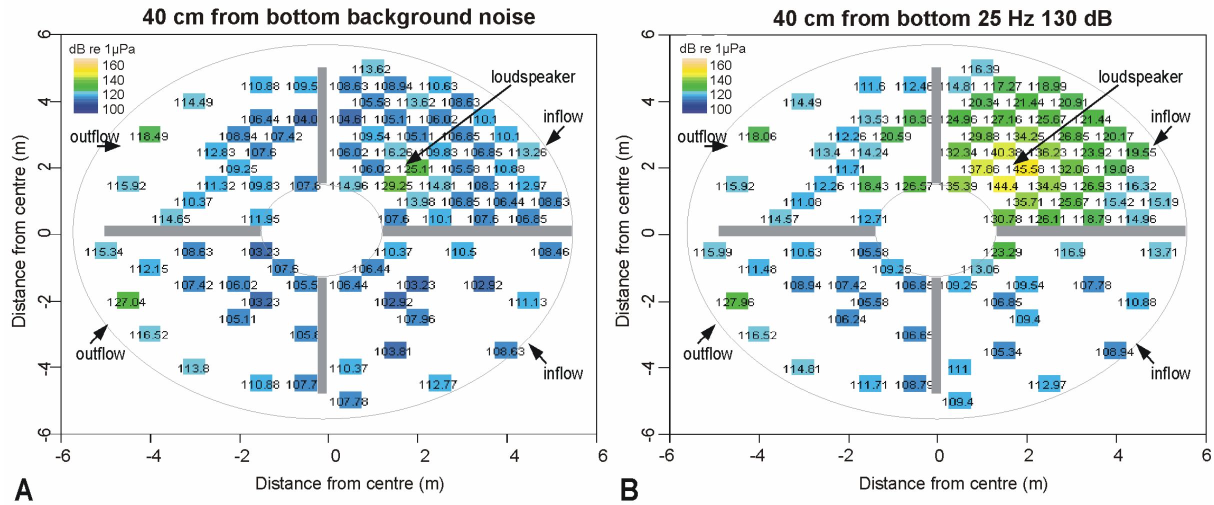


Fig. 29: Comparison of background sound field (**A**) and sound field during production of 25 Hz at 130 dB re 1µPa 0.4 m above the bottom of the tank (**B**). The measurements are displayed as absolute readings. Local high readings in the background noise were caused by the turned off loudspeaker in quarter 1 and the water outflows in quarter 2 and 3. Fig. B is the same as Fig. 24 but absolute sound pressure levels.

Increased background noise was caused by the water outflows and surprisingly by the switched off loudspeaker 1. The readings next to the loudspeaker close to the bottom and 0.4 m above the tank bottom reached up to 130 dB re 1µPa which was much higher than the surrounding sound levels. The higher sound levels were only local not affecting the overall sound field in quarter 1. This effect could be seen in the measurements with sound barriers as well as in the later following measurements without the sound barriers in place. Caused by the local high readings a sound pressure difference in the tank existed but was not suitable to describe the background noise field in the tank.

The average sound levels of the quarters 2-4 during sound production were compared with the average background noise level in the same area of the tank (Table 6). The results show a higher sound level in all tested sound situations compared with the background noise level in the quarters 2-4.

During the experiments at a sound level of 130 dB re 1µPa the sound level decreased to the level of the background noise in small areas of the tank. At the sound level of 140 dB re 1µPa the sound levels exceeded the background noise levels but there were also areas with distinctly lower sound levels.

Table 6: Differences between average produced sound levels and average background noise levels in quarters 2-4 in the divided tank in dB

	25 Hz 130 dB	60 Hz 130 dB	90 Hz 130 dB	125 Hz 130 dB	250 Hz 130 dB	60 Hz 140 dB	90 Hz 140 dB	125 Hz 140 dB	250 Hz 140 dB
bottom	3.46	7.06	5.89	5.43	8.26	12.72	12.29	10.54	15.06
40 cm from b.	3.13	6.98	5.74	5.71	7.33	13.43	12.55	10.66	13,96
80 cm from b.	2.83	7.02	5.62	6.01	7.43	13.03	11.87	11.14	14.12
surface	2.22	5.19	4.04	4.50	6.80	11.12	9.24	9.28	12.09

The table contains the differences [dB] between the average sound levels in different produced sound fields and the average background noise level for the combined quarters 2-4 in the tank on the basis of 49 measurement points.

4.1.2 Acoustic field without sound barriers

For comparison, the sound level was measured in the tank with the barriers removed. For technical reasons, this was done after the experiments. To examine the influence of the sound barriers on the acoustic field in the tank, measurements were carried out at 29 points in the tank after removing the barriers (Fig. 30). The 29 measurement points were a selection of those 86 points used in the previous measurements with barriers in place (Fig. 23). Measurements were done at four water depths and loudspeaker 1 in quarter 1 was used for sound production. After the long period of time under water the loudspeaker was weaker than it was in the earlier measurements. The frequency 25 Hz could only be produced at a sound level of 121 dB re 1µPa, the sound production at 60 Hz reached only 130 dB re 1µPa and at 90 Hz 137 dB re 1µPa was the maximum sound level.

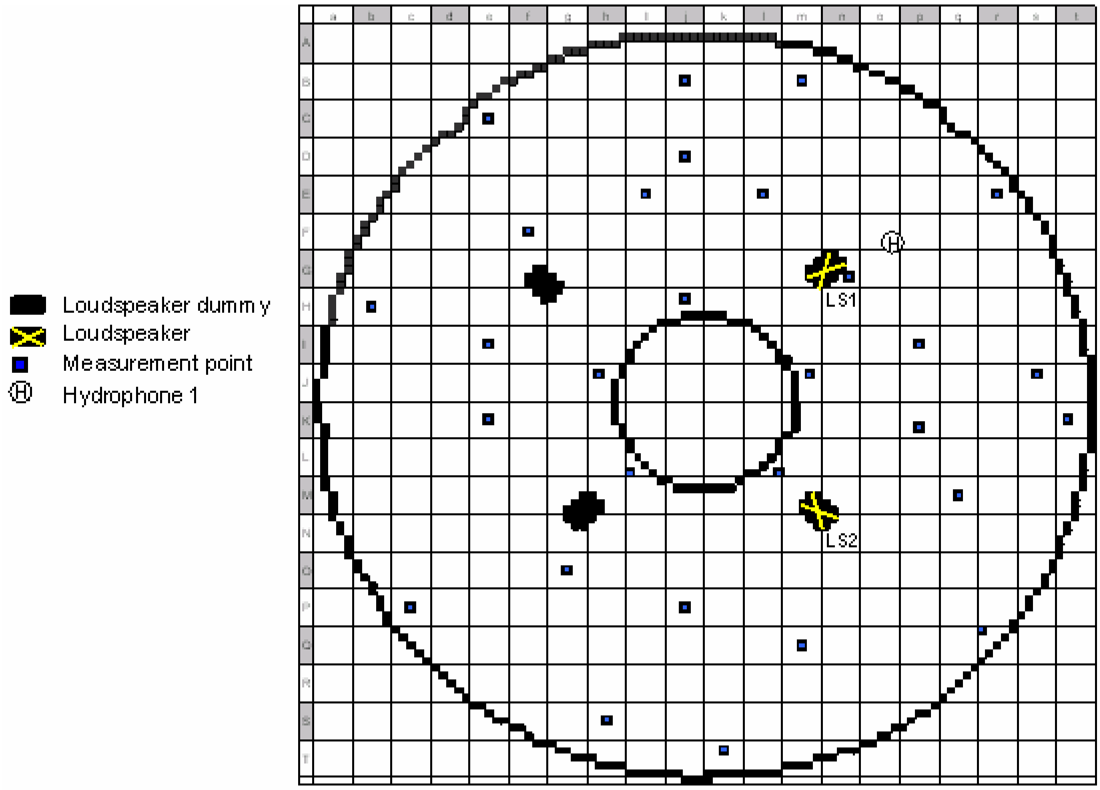


Fig. 30: Position of measurement points in the experimental tank without sound barriers in place. The sound was produced by loudspeaker 1 in quarter 1. The measurements were made at 29 measurement points at four different water depths.

Fig. 31 shows the results of measurements with and without sound barriers for the frequencies 90 Hz at 130 dB re 1µPa and 250 Hz at 140 dB re 1µPa. Most pronounced differences between the sound fields appeared at the frequency of 250 Hz with the effect decreasing with frequency. At 250 Hz the maximal sound level decrease in the tank without barriers was -18 dB while it was more than –31 dB in the divided tank. At 90 Hz the sound level decrease in the tank without barriers in place was only slightly smaller than in the divided tank. Especially in the vicinity of the observation tube in the middle of the tank the decrease was less since the sound could travel around the tube without being hold back by the barriers. The data for other frequencies and intensities are given in the appendix (Fig. A 18 to Fig. A 25).

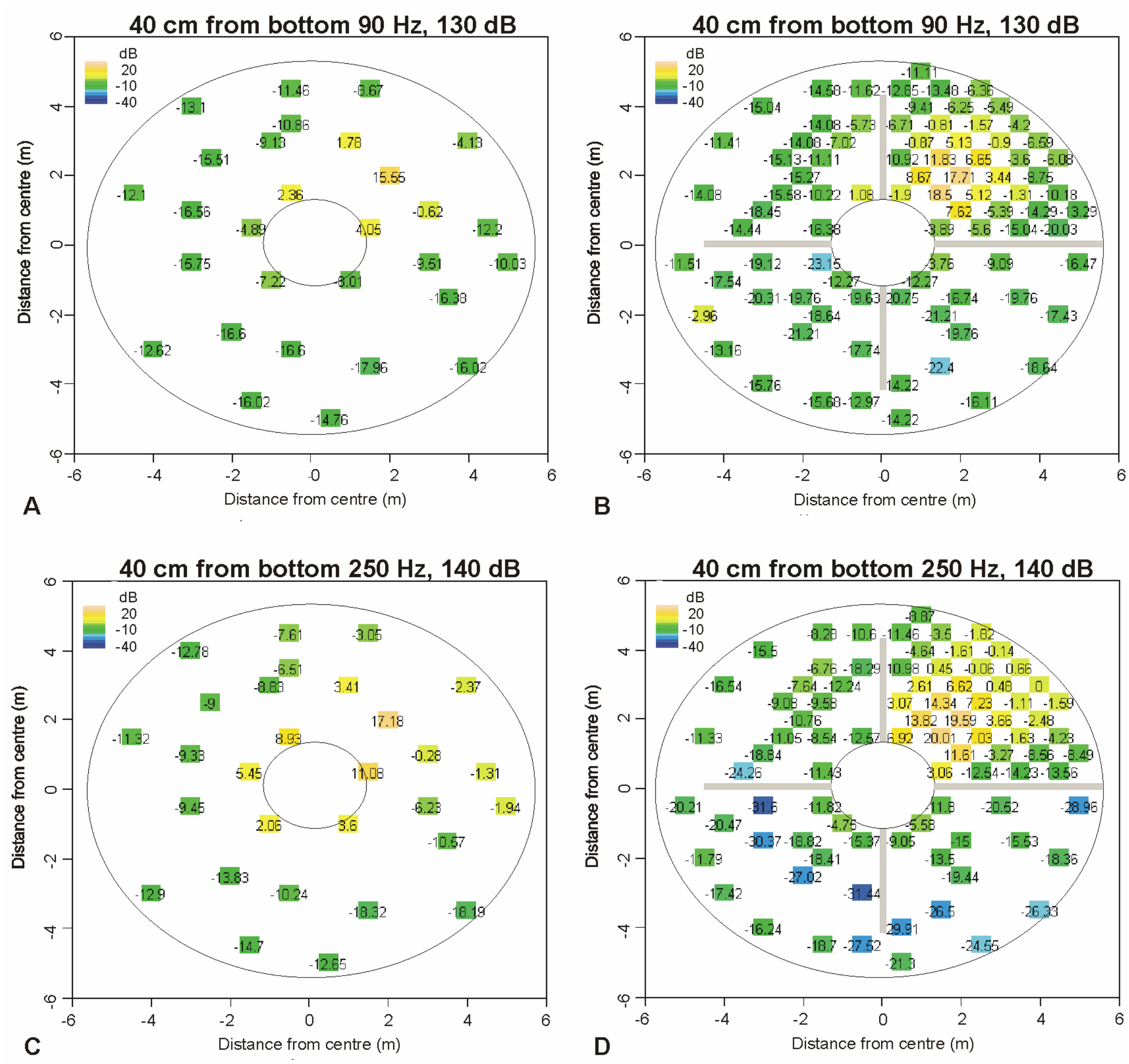


Fig. 31: Comparison between the sound fields with and without barriers during sound production of 90 Hz (130 dB re 1µPa) and 250 Hz (140 dB re 1µPa) 0.4 m above the bottom of the tank. Sound levels are displayed as the difference from the reference sound level at hydrophone H1. **A+B**: At 90 Hz the differences appeared mostly in the vicinity off the observation tube and in the areas where the barriers separated quarter 1 from the neighbouring quarters. **C+D**: At the frequency of 250 Hz (140 dB re 1µPa) areas of high sound level decrease are visible by blue colours in quarters 3 and 4 of the divided tank. In comparison the sound level decrease in these areas could be more than 20 dB smaller in the undivided tank than in the tank with barriers. Also obvious was the spreading of the sound around the observation tube in the middle of the undivided tank.

To compare the sound fields with and without barriers the maximal sound pressure differences for given stimulus conditions in the divided tank were determined on the base of the 29 measurement points used in the measurements without barriers. The maximal differences in the tank with barriers were between 31 and 52 dB (Table A 3). The experiment 25 Hz at 130 dB re 1µPa was excluded from this calculation because this sound level could not be produced in the experiments without barriers due to a weakness of the loudspeaker at low frequencies. Without barriers the maximal sound pressure differences were much smaller being between 25 and 42 dB (Table A 4). The differences during different sound stimuli could come up to nearly 18 dB. The sound pressure differences in the tank with and without barriers during different stimuli are displayed in Fig. 32 and Table 7 showing an increasing sound pressure difference and therefore increasing effect of the barriers with frequency.

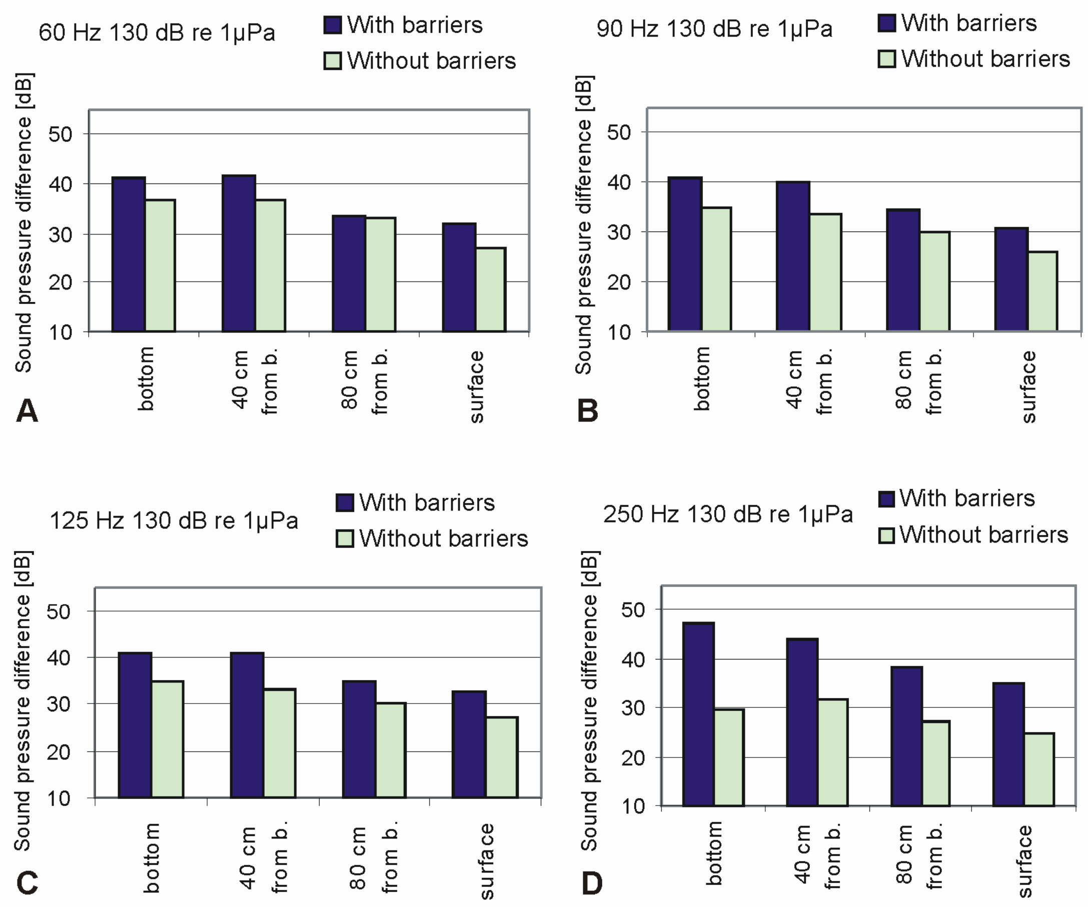


Fig. 32: Comparison of sound pressure differences in the tank with and without barriers during sound production of 130 dB re 1µPa at different water depths. **A**: At a frequency of 60 Hz the pressure difference at the different water depths got up to 5 dB. **B**: At 90 Hz the sound pressure difference reached up to 6.6 dB. **C**: At 125 Hz the difference reached up to 7.6 dB. **D**: At a frequency of 250 Hz the sound pressure differences between 10 dB and nearly 18 dB were much higher compared with the lower frequencies.

Table 7: Sound pressure differences in the tank with and without barriers

sound stimulus	Sound pressure differences [dB]							
	bottom		40 cm		80 cm		surface	
	with barriers	without barriers	with barriers	without barriers	with barriers	without barriers	with barriers	without barriers
60 Hz, 130 dB	41,11	36,68	41,73	36,80	33,56	33,29	31,92	27,04
90 Hz, 130 dB	41,10	35,11	40,11	33,51	34,65	30,12	30,95	26,14
125 Hz, 130 dB	41,06	34,70	40,68	33,11	34,81	30,32	32,51	27,04
250 Hz, 130 dB	47,40	29,75	43,77	31,52	38,43	27,07	34,86	24,81
125 Hz, 140 dB	45,45	41,78	48,30	41,14	41,28	36,62	39,62	31,72
250 Hz, 140 dB	48,75	35,40	51,64	35,50	47,05	31,48	43,52	28,05

Difference of minimal and maximal readings during different sound stimuli in the tank with and without barriers at four water depths. The values are based on 29 measurement points.

While Fig. 31 and Fig. 32 compare the sound field in the whole tank the following evaluations exclude the quarter of sound production to compare only the sound fields in the quarters 2-4 that were separated from the sound source by barriers in the divided tank.

The barriers left a gap of about 70 cm at the outer wall of the tank. Therefore higher sound levels next to the outer wall were expected where the sound waves could propagate undisturbed by barriers. Sound level differences could be expected from the outer wall towards the inner, more protected areas of the tank. For that reason the measurement points used for the comparison of the sound fields with and without barriers were separated in four circles of about 90 cm radius each. Fig. 33 shows the position of measurement points in quarter 2-4 arranged in four circles.

In the divided tank the average sound levels at the lower frequencies of 60 Hz and 90 Hz hardly differed between circles (Fig. 34A,B). At the frequencies of 125 Hz and 250 Hz the sound levels increased clearly in circle 4 close to the observation tube (Fig. 34C,D). In the tank without barriers the central circles 2 and 3 showed the lowest sound levels. A strong increase in sound levels could be seen in the inner circle 4.

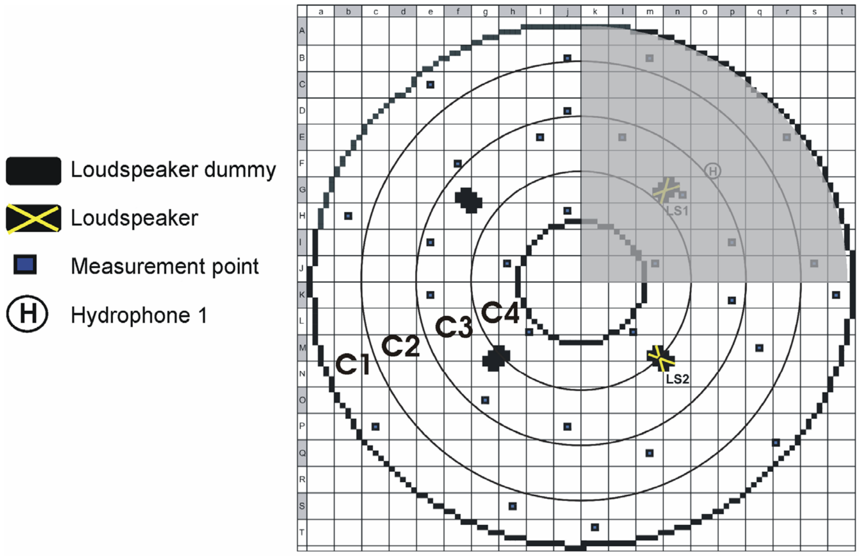


Fig. 33: Position of circles for annular sound pressure level evaluation. The measurement points in the quarter of sound production (covered in grey) are excluded from the evaluation.

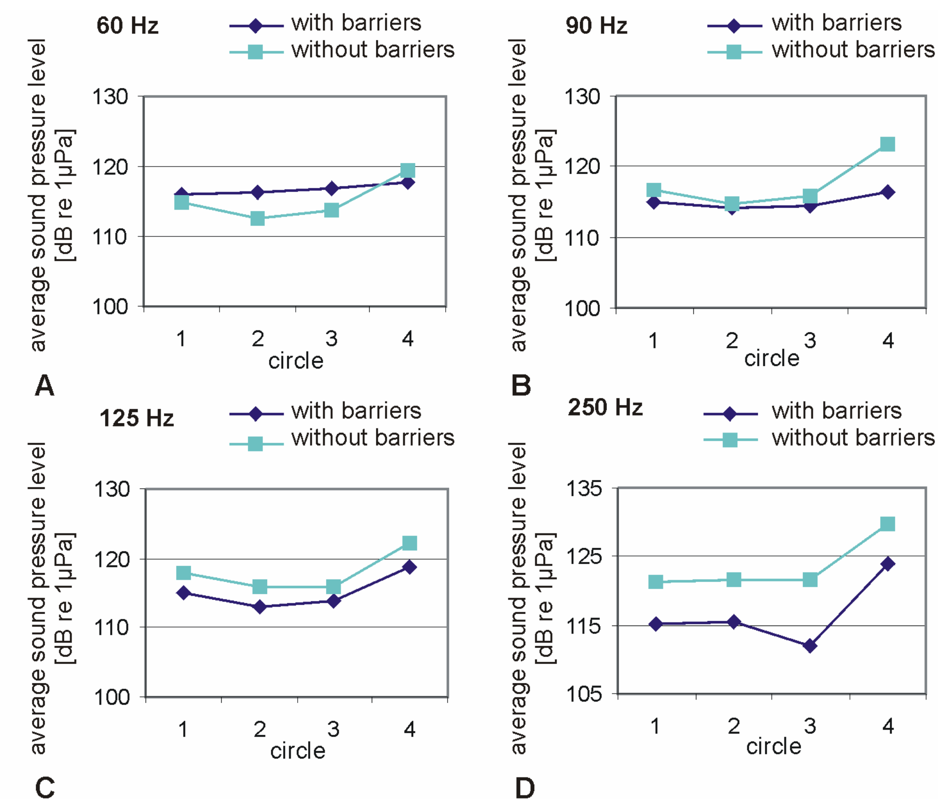


Fig. 34: Average sound pressure levels in tank quarter 2-4 with and without barriers during sound production of 130 dB re 1µPa. The measurement points are arranged in four circles of about 90 cm radius, starting with circle 1 at the outer wall to circle 4 next to the observation tube in the middle of the tank. **A**: 60 Hz **B**: 90 Hz **C**: 125 Hz **D**: 250 Hz.

From the different evaluations an effect of the barriers on the acoustic field in the tank could be demonstrated from a frequency of 90 Hz. The effect increased with the frequency and was most obvious at 250 Hz.

4.2 Behavioural experiments

4.2.1 Experiments using cod

4.2.1.1 Observations without sound production

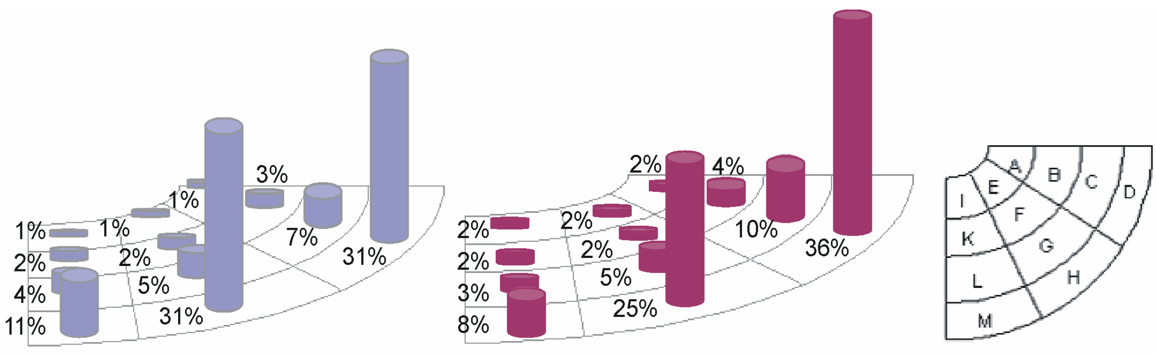
4.2.1.1.1 General behavioural

On average about two thirds of juvenile and adult cod present in tank quarter 4 were swimming while the other third rested. Fish rested mainly facing into the water inflow but also in the water column or on sand bags positioned along the sound barriers. Swimming was observed in the whole water column and fish frequently formed small schools.

4.2.1.1.2 Distribution of cod in the tank

Adult cod showed a preference for tank quarter 4 where on average 38% to 92% of the fish could be observed. Apart from the first experiment juvenile cod also showed a preference for quarter 4 with an average of 43% to 64% found there during experiments. In most cases the number of adult cod in quarter 4 was higher than it was in juveniles. In the first week following release of juvenile cod into the tank fish were evenly spread between quarters 1 and 4 with about 40% present in each.

The distribution in quarter 4 showed a strong preference for sections D and H next to the water inflow, where most of the resting took place. Combining the data from all experiments 64% of the juvenile and 62% of the adult cod present in quarter 4 were found in these sections (Fig. 35).



A B C

Fig. 35: Distribution of juvenile (**A**) and adult (**B**) cod displayed as the percentage of fish present in quarter 4. Data from periods without sound production pooled for all experiments. **C**: Position of sections in quarter 4.

From the results it can be seen that cod used the outer sections of the tank more often than the inner sections next to the observation chamber.

4.2.1.1.3 Diel rhythm in cod

The number of both juvenile and adult cod in quarter 4 was significantly higher in daytime than it was at night in most of the experiments. An example is given in Fig. 36.

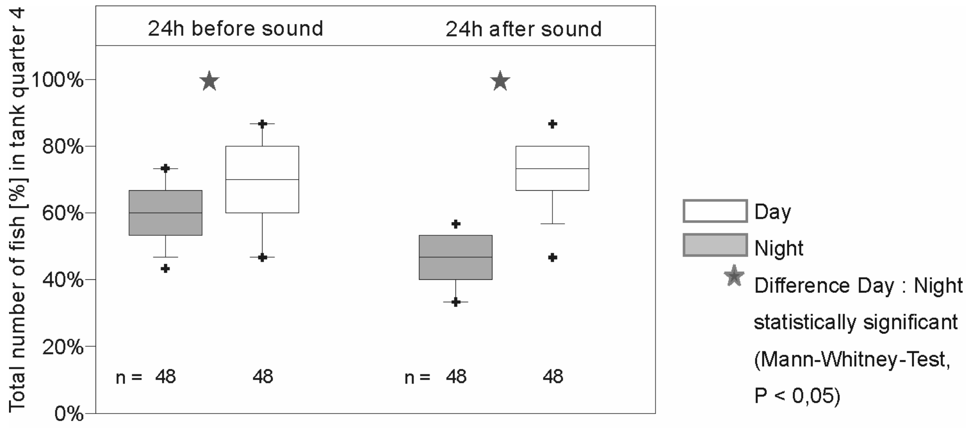


Fig. 36: Percentage of juvenile cod in quarter 4 of all fish in the tank at day and night shown for two days as an example. Significant differences between day and night are marked by an asterisk above the Box-Whisker-Plots. n = number of pictures evaluated.

Usually cod were more active in daytime than at night, when the number of resting fish in quarter 4 increased (Fig. 37). This was more pronounced in juvenile than in adult cod. Higher activity in daytime was observed in 23 of the 24 days testing juvenile cod and 24 of the 27 days testing adult cod. An example is given in Fig. 38. The level of activity varied strongly between days with average activity levels of between less than 30% and more than 90%. Statistical results can be found in Table A 5 (appendix).

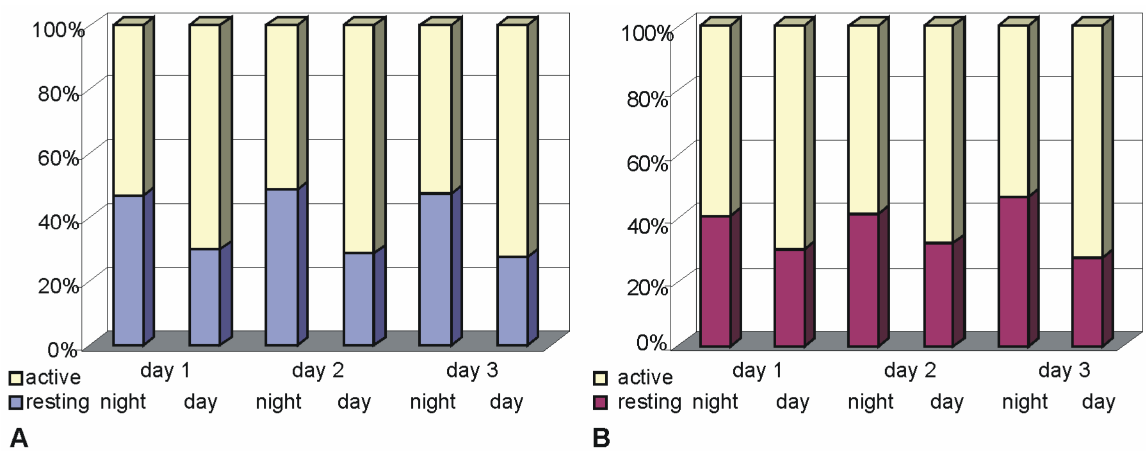


Fig. 37: Percentage of activity and resting behaviour (median) of juvenile (**A**) and adult (**B**) cod present in quarter 4 during a 72 hours observation period. The figures contain the pooled results of all experiments with sound production in quarter 4.

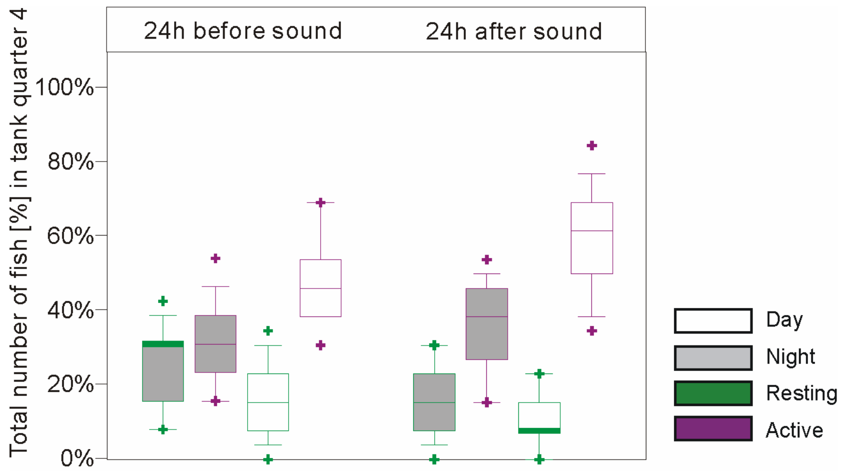


Fig. 38: Percentage of adult cod active or resting at day or night shown for two days as an example

4.2.1.1.4 Feeding

Although cod preferred quarter 4, they also frequently moved around other parts of the tank. The food provided was found and eaten by the fish within a few minutes, so the feeding event did not cause obvious changes in distribution in most of the experiments. Only when the data were displayed in fine detail feeding effects are evident in some experiments. Examples of experiments with and without feeding effects using the same group of fish are given in Fig. 39A and B.

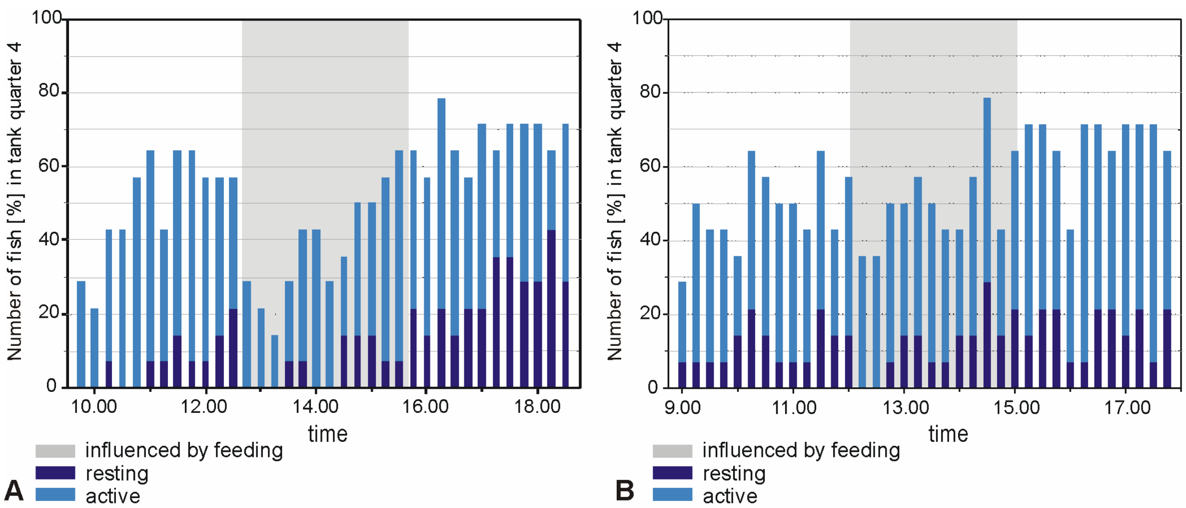


Fig. 39: Number and behaviour of juvenile cod in quarter 4 three hours before and six hours after a feeding. A period of three hours after feeding is coloured. **A**: Decreasing number of fish in quarter 4 after feeding. **B**: The number of fish remained nearly unchanged in quarter 4 after feeding.

4.2.1.2 Behaviour of cod during sound production

The results of the experiments using cod are displayed as box plots showing the fish numbers present in quarter 4 in the 24-h-periods before, during and after sound production (Fig. 40 to Fig. 44). Additionally the average fish numbers (median) present in the periods before, during and after sound and the p-values of the Mann-Whitney-Test with Bonferroni-correction for significant differences between the different periods are given in Table 8 to Table 12. Additional figures showing the overall fish numbers present in quarter 4, the fish numbers divided into active and resting and box plots showing the fish numbers in the periods before, during and after sound at day and night as well as divided in active and resting can be found in Fig. A 26 to Fig. A 44 (appendix). All statistical results of the Kruskal-Wallis-test and the Mann-Whitney-Test are summarized in Table A 6 (appendix).

4.2.1.2.1 Tested frequency of 25 Hz

In juvenile cod significant reactions to the sound of 25 Hz were observed only at the higher sound level of 140 dB re 1µPa with lower fish numbers during sound production compared with the periods before and after sound. The same reaction was observed in adult cod at 130 dB re 1µPa while the fish numbers remained on the same level during sound production of 140 dB re 1µPa but increased significantly after the sound was switched off. A significant difference also existed between the periods before sound and after sound with lower fish numbers before the sound was turned on.

Table 8: Average fish numbers (median) present in quarter 4 in experiments using cod during sound production of 25 Hz.

Juvenile cod	Before sound	During sound	After sound
25 Hz, 130 dB re 1µPa	42.86%	35.71%	35.71%
	p = 0.0000	p = 0.0000	
25 Hz, 140 dB re 1µPa	50.00%	35.7%	50.00%
Adult cod	**Before sound**	**During sound**	**After sound**
	p = 0.0000	p = 0.0000	
25 Hz, 130 dB re 1µPa	61.54%	50.00%	61.54%
		p = 0.0000	
25 Hz, 140 dB re 1µPa	69.23%	69.23%	76.92%
		p = 0.0000	

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

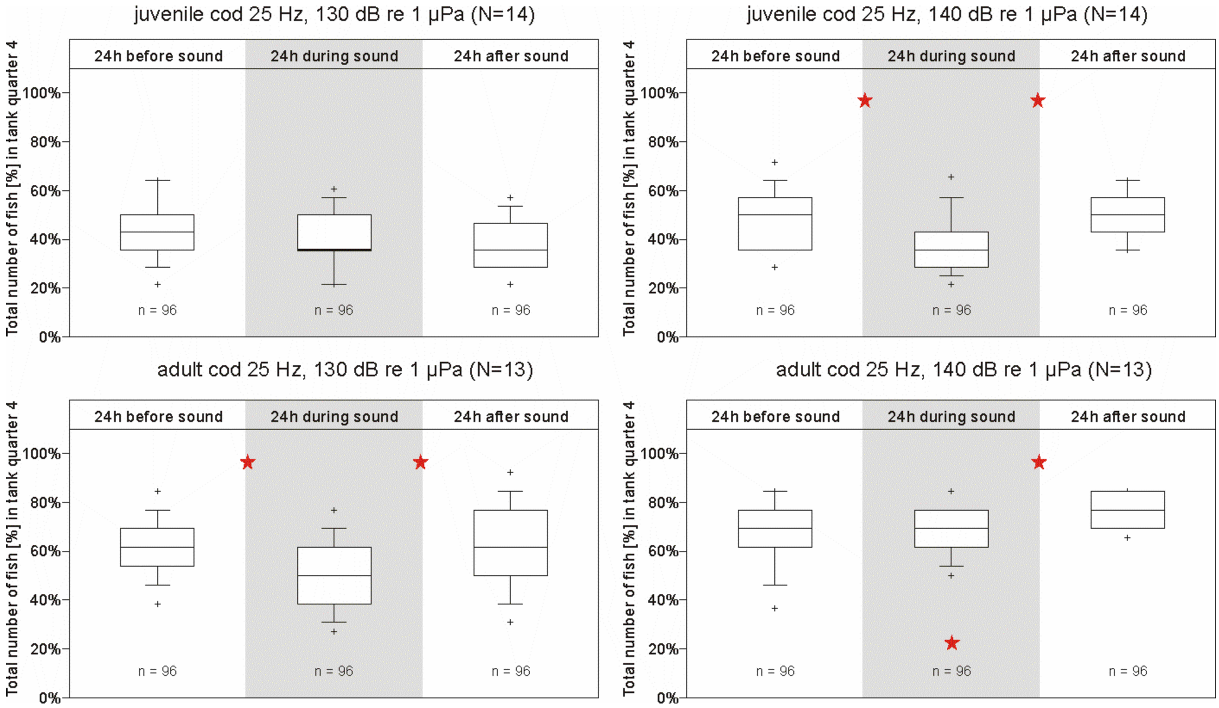


Fig. 40: Number of juvenile and adult cod (median) present in quarter 4 before, during and after sound production of 25 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

4.2.1.2.2 Tested frequency of 60 Hz

In both experiments using juvenile cod at a frequency of 60 Hz the fish numbers decreased significantly during sound production. While the fish numbers returned to pre-noise level at 140 dB re 1 µPa they increased but remained on a significantly lower level compared with the fish numbers before sound production of 130 dB re 1µPa. The reaction of juvenile cod to sound of 60 Hz at 140 dB re 1µPa was strong with a decrease by 29% compared with the periods without sound.

At the sound level of 130 dB re 1µPa the number of adult cod in quarter 4 decreased during sound production but the difference was not significant. Significant differences appeared between the period before sound and the during and after sound periods. At the higher sound level the fish numbers were significantly lower in the sound period compared with the periods before and after sound.

Table 9: Average fish numbers (median) present in quarter 4 in experiments using cod during sound production of 60 Hz.

Juvenile cod	Before sound	During sound	After sound
	p = 0.0000		p = 0.0001
60 Hz, 130 dB re 1µPa	66.67%	46.67%	53.33%
		p = 0.0038	
	p = 0.0000		p = 0.0000
60 Hz, 140 dB re 1µPa	71.43%	42.86%	71.43%
Adult cod	**Before sound**	**During sound**	**After sound**
			p = 0.0000
60 Hz, 130 dB re 1µPa	84.62%	76.92%	92.31%
		p = 0.0000	
	p = 0.0000		p = 0.0000
60 Hz, 140 dB re 1µPa	61.54%	38.46	61.54

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

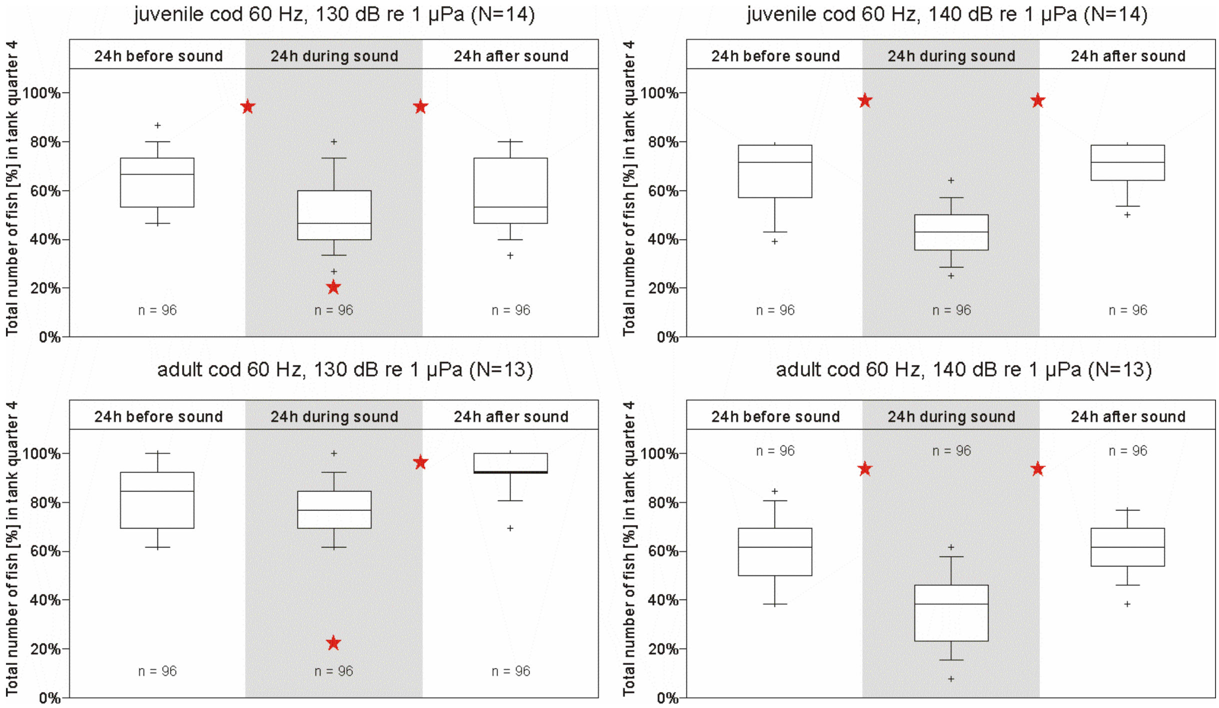


Fig. 41: Number of juvenile and adult cod (median) present in quarter 4 before, during and after sound production of 60 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

4.2.1.2.3 Tested frequency of 90 Hz

Sound at a frequency of 90 Hz at 130 dB re 1µPa did not produce a significant reaction in juvenile cod although the average fish numbers in quarter 4 increased significantly after the sound was turned off compared with the periods before and during sound production.

The experiment with sound production of 90 Hz at 140 dB re 1µPa differed fundamentally from all other experiments as it was carried out using the loudspeaker in quarter 1 instead of quarter 4. In the period before sound production the average number of fish present in quarter 1 was 40%. After the sound was turned on the fish numbers dropped significantly to about 13% and increased again to 20% in the period after sound production. The fish leaving quarter 1 appeared to enter quarter 4 where the proportion increased from 40% before to more than 73% during sound production and decreased to 60% after the sound was switched off. After this experiment the fish numbers in quarter 1 remained low for the rest of the experimental series. The observed differences between the three 24h periods were all significant both in quarter 1 and 4.

Table 10: Average fish numbers (median) present in quarter 4 in experiments using cod during sound production of 90 Hz.

Juvenile cod	Before sound	During sound	After sound
			p = 0.0000
90 Hz, 130 dB re 1µPa	42.86%	42.86%	50.00%
		p = 0.0000	
	p = 0.0000		p = 0.0000
90 Hz, 140 dB re 1µPa Quarter 1	40.00	13.33%	20.00%
		p = 0.0000	
	p = 0.0000		p = 0.0000
90 Hz, 140 dB re 1µPa Quarter 4	40.00%	73.33%	60.00%
		p = 0.0000	
Adult cod	**Before sound**	**During sound**	**After sound**
	p = 0.0000		p = 0.0000
90 Hz, 130 dB re 1µPa	69.23%	53.85%	73.08%
	p = 0.0000		
90 Hz, 140 dB re 1µPa	57.69%	38.46%	38.46%
		p = 0.0000	

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

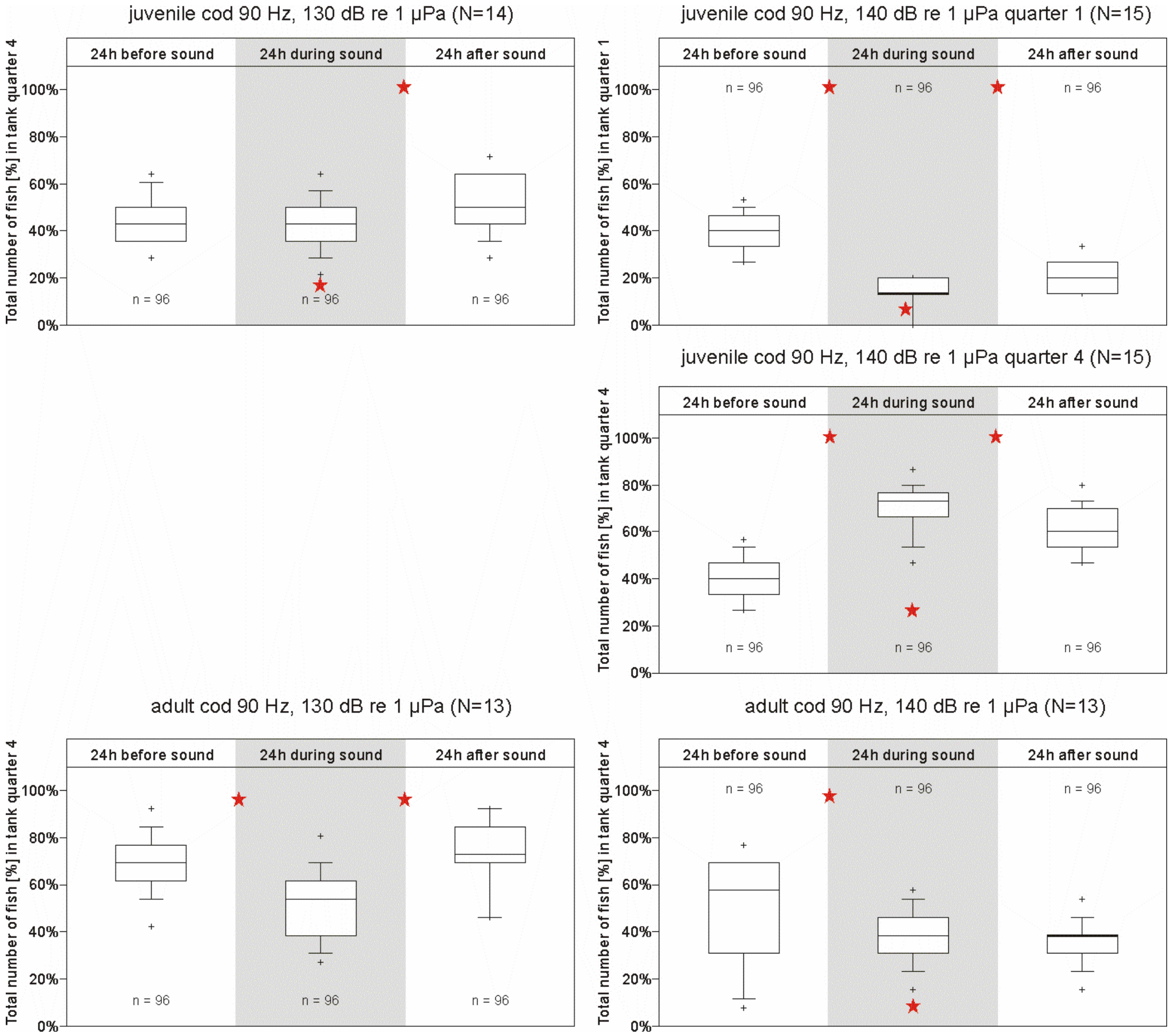


Fig. 42: Number of juvenile and adult cod (median) present in quarter 4 and quarter 1 before, during and after sound production of 90 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

In the experiments using adult cod the fish numbers decreased significantly during sound production at both sound levels. While the proportion of fish returned to a even higher level after sound of 130 dB re 1µPa it remained on a low level after the sound of 140 dB re 1 µPa was switched off. However the result of this experiment was influenced by a reaction that was not related to sound production since it occurred in the early morning, before turning the sound on in the afternoon. Shortly after the light came on, the fish numbers in quarter 4 decreased suddenly from between 60% to 70% to less than 10% and increased only very slowly over the following hours. The reason for this sudden reaction was not determined.

4.2.1.2.4 Tested frequency of 125 Hz

During sound production of 125 Hz at 130 dB re 1µPa the number of juvenile cod decreased significantly and increased again to a lower level than before sound but this difference was also significant. At 140 dB re 1µPa the median proportion of juvenile cod in quarter 4 remained the same, however the variation in numbers around that median indicated a significant decrease once the sound of 125 Hz was switched on.

At 130 dB re 1µPa, adult cod did not show any reaction to the sound. Over the whole 72 hours period of this experiment the average fish numbers present in quarter 4 remained at more than 92% with an unusually high amount of resting fish (46% to 62%). At 140 dB re 1µPa the average number of fish was significantly lower during sound production compared with the periods before and after sound.

Table 11: Average fish numbers (median) present in quarter 4 in experiments using cod during sound production of 125 Hz

Juvenile cod	Before sound	During sound	After sound
	p = 0.0000		p = 0.0001
125 Hz, 130 dB re 1µPa	64.29%	50.00%	57.14%
		p = 0.0001	
	p = 0.0123		p = 0.0000
125 Hz, 140 dB re1µPa	50.00%	50.00%	60.71%
		p = 0.0000	
Adult cod	**Before sound**	**During sound**	**After sound**
125 Hz, 130 dB re 1µPa	92.31%	92.31%	92.31%
	p = 0.0000		p = 0.0007
125 Hz, 140 dB re 1µPa	76.92%	61.54%	69.23%
		p = 0.0013	

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

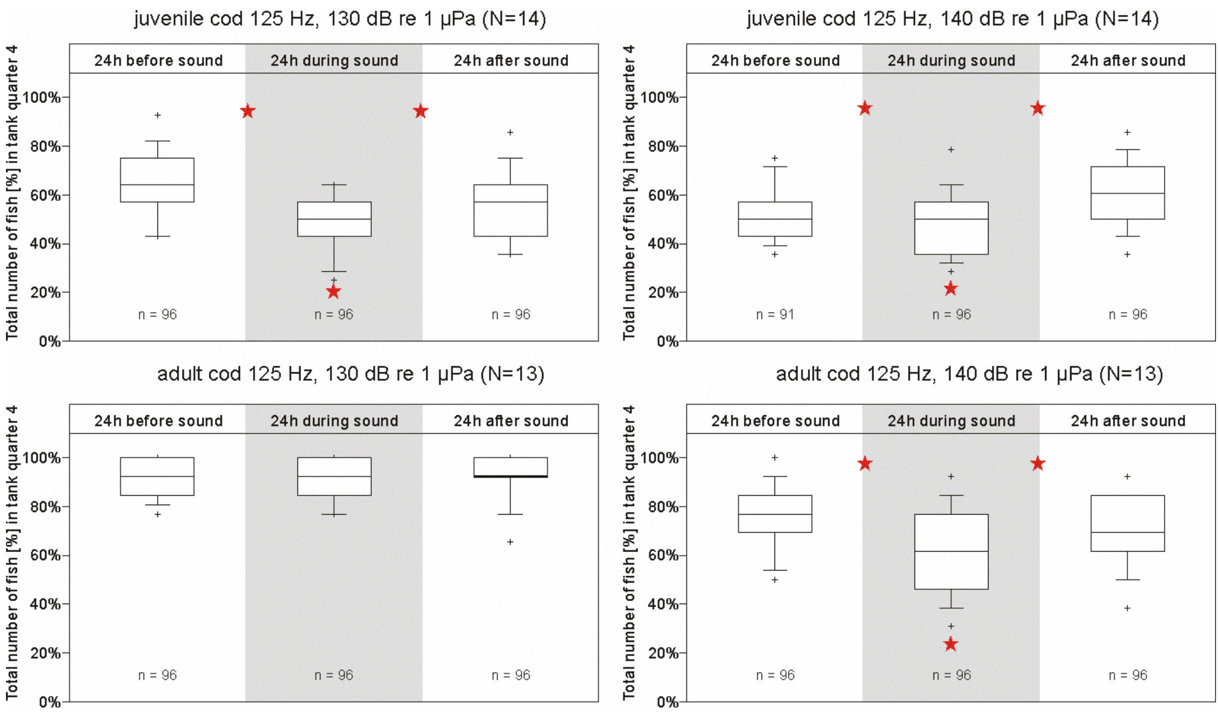


Fig. 43: Number of juvenile and adult cod (median) present in quarter 4 before, during and after sound production of 125 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

4.2.1.2.5 Tested frequency of 250 Hz

The experiments using the frequency of 250 Hz were only carried out at a sound level of 140 dB re 1µPa.

The average number of juvenile cod present in quarter 4 decreased only slightly during sound production, and increased significantly after the sound was switched off. In adult cod no significant avoidance of quarter 4 was observed during sound production.

Table 12: Average fish numbers (median) present in quarter 4 in experiments using cod during sound production of 250 Hz

Juvenile cod	Before sound	During sound	After sound
		p = 0.0000	
250 Hz, 140 dB re 1µPa	46.43%	42.86%	50.00%
Adult cod	**Before sound**	**During sound**	**After sound**
250 Hz, 140 dB re 1µPa	69.23%	61.54%	61.54%

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

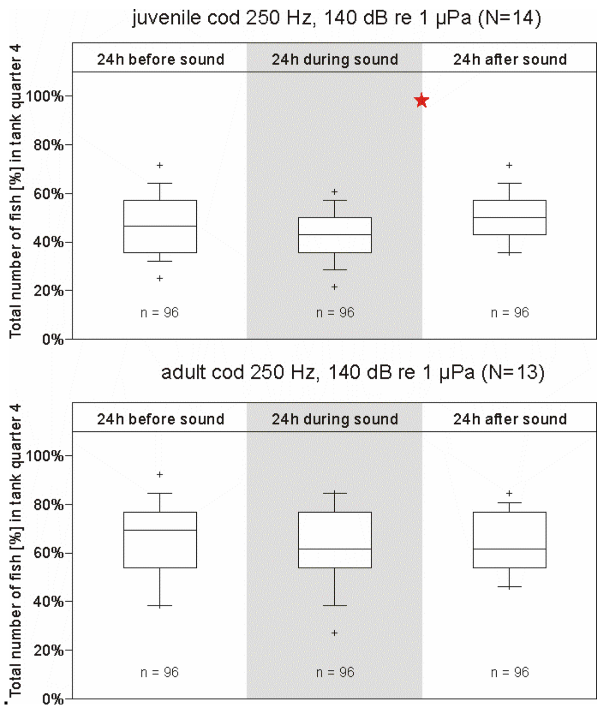


Fig. 44: Number of juvenile and adult cod (median) present in quarter 4 before, during and after sound production of 250 Hz at a sound level of 140 dB re 1µPa. N = fish numbers present in the tank, n = number of pictures evaluated. The significant differences between 24 hours periods is marked with asterisks.

4.2.1.2.6 Loudspeaker vicinity evaluation

In many experiments using cod a significant avoidance of quarter 4 during sound production was observed, although in some experiments, the fish numbers remained approximately the same.

The sound levels in tank quarter 4 declined steeply from up to about 161 dB re 1µPa in the direct vicinity of the sound source to 17 to 41 dB lower levels around the edges of the quarter depending on frequency, sound level and water depth. Therefore a movement away from the highest sound levels would be possible without leaving quarter 4. Two experiments using juvenile and two with adult cod were chosen from the sound experiments for more detailed evaluation. While one experiment each in juvenile and adult cod showed significantly lower fish numbers in quarter 4 during sound production in the earlier evaluation of quarter 4 in the other experiment the fish numbers remained on about the same level while the sound was turned on.

The evaluation of a circle 1 m around the loudspeaker centre showed lower number of fish sightings within 1 m distance during sound production than in the periods before and after sound in all four experiments (Fig. 45). The number of adult cod entering the circle during a 30 minute period was higher than it was in juvenile cod which reflects the higher numbers of adult cod present in quarter 4 compared with juvenile cod.

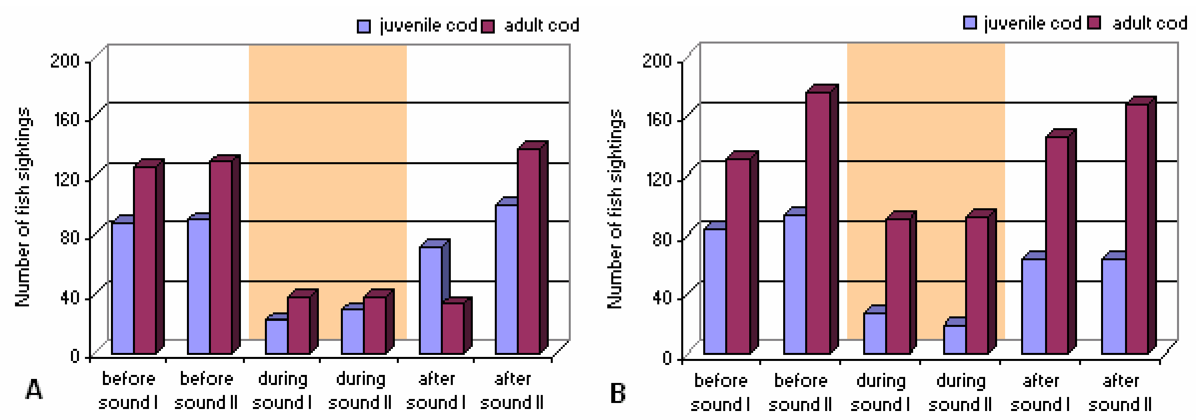


Fig. 45: Number of sightings of fish entering a circle of one meter around the loudspeaker in 30 minute periods at the beginning and the end of the phases before, during and after sound production each. The sound period is coloured. **A**: Experiments with juvenile (60 Hz, 130 dB) and adult cod (90 Hz, 130 dB) showing significantly lower fish numbers while in **B** only small or no changes in fish numbers in quarter 4 during sound production in the overall evaluation were observed (experiments juvenile cod 25 Hz, 130 dB, adult cod 125 Hz, 130 dB).

When counting fish sightings the duration of every sighting from entering to leaving the circle was determined (Fig. 46). Overall the presence within 1 m was mostly brief with 37% to 95% of the sightings being 10 or less seconds in juveniles and about 70% to 100% in adult cod. A tendency to shorter stays during sound production was observed in the experiments.

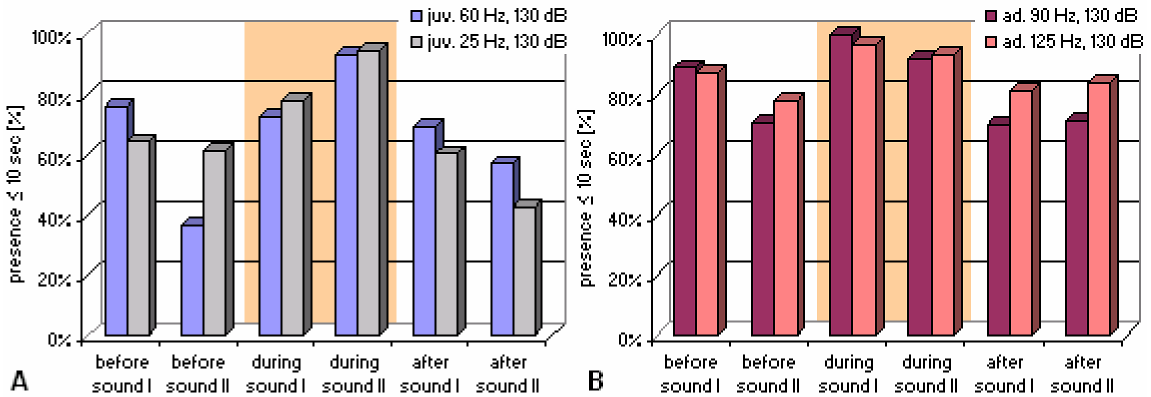


Fig. 46: Percentage of short duration presence of 10 or less seconds in a circle of one meter around the loudspeaker in 30 minute periods at the beginning and the end of the phases before, during and after sound production each. The sound period is coloured. **A**: Experiments using juvenile cod. **B**: Experiments using adult cod.

As already seen from the percentage of short stays within 1 m of the loudspeaker, juvenile cod stayed longer than adults. The average duration (median) of presence is displayed in Fig. 47 showing longer stays in most phases within juvenile cod.

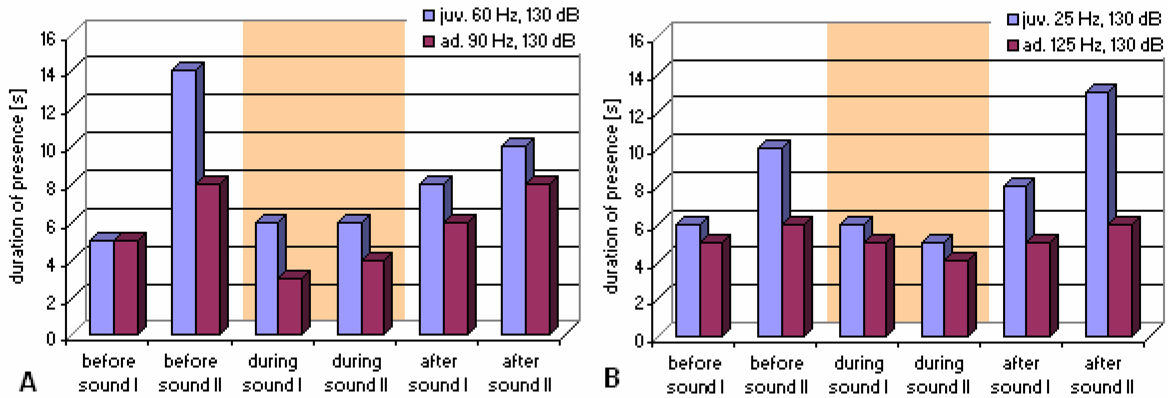


Fig. 47: Average duration of presence (median) in a circle of one meter around the loudspeaker in 30 minute periods at the beginning and the end of the phases before, during and after sound production each. The sound period is coloured. **A**: Experiments with and **B** without significant lower fish numbers in quarter 4 during sound production in the overall evaluation.

The results of the vicinity evaluation showed lower fish numbers in the vicinity of the loudspeaker during sound production even in experiments with no significant differences in fish numbers between the different periods in quarter 4. Looking at the distribution of fish in the sections of quarter 4 a shift to the outer sections could be seen during sound production. As an example, for an experiment with significantly lower fish numbers during sound production the distribution of fish in tank quarter 4 is given for the experiment of 60 Hz at 130 dB re 1µPa using juvenile cod (Fig. 48A-C). Decreasing fish numbers became obvious in the inner sections of the tank but also in the preferred section D. In experiments without significantly changing fish numbers in quarter 4 the shift of fish away from the sound source led to increasing fish numbers in the sections D and H (Fig. 48D-F).

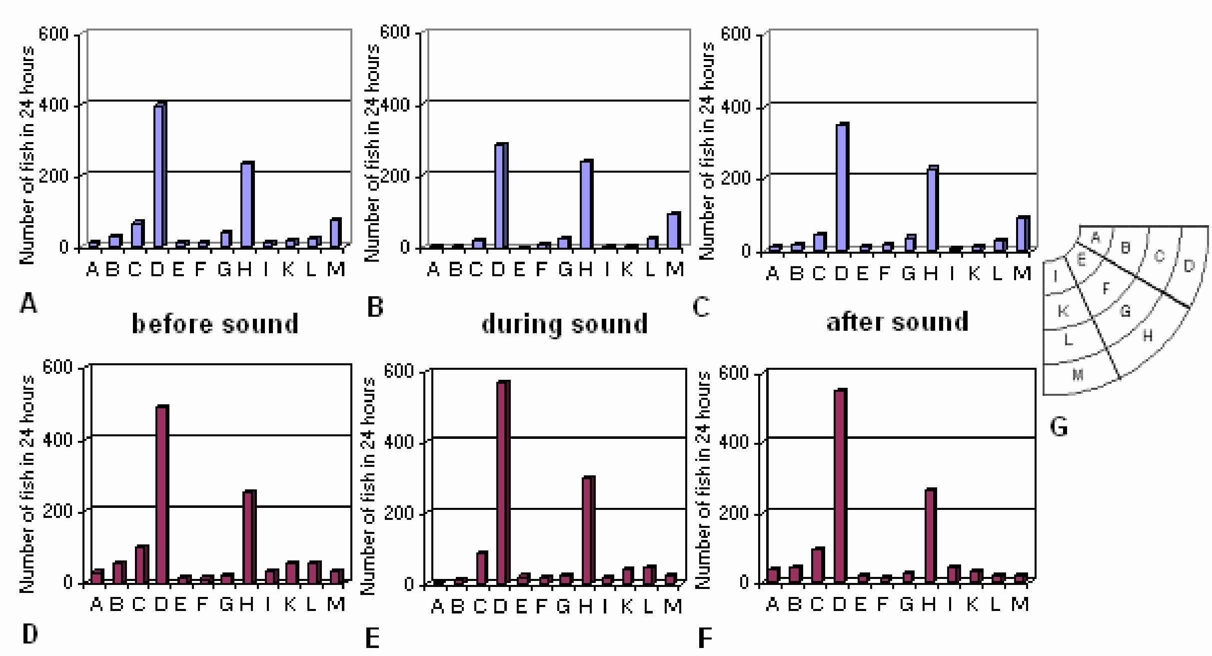


Fig. 48: Number of cod present in the sections of quarter 4 in the 24 hour periods before, during and after sound. **A-C**: Juvenile cod (experiment 60 Hz, 130 dB). A decrease of fish numbers during sound production could be seen in the inner sections as well as in the preferred section D that led to significant lower fish numbers in quarter 4. **D-F**: Adult cod (experiment 125 Hz, 130 dB). A shift of fish from the inner sections of the tank to the outer sections D and H could be seen during sound production while the fish numbers in the quarter remained stable. **G**: Position of sections.

4.2.1.3 Recapitulation of cod results

To summarize the results of cod the difference of the arithmetic mean of the fish numbers present in quarter 4 in the periods before and during sound production are displayed in Fig. 49. Significant avoidance reactions to low frequency sound between 25 and 125 Hz were observed, with the strongest reactions to 60 and 90 Hz. From the circumference evaluation an effect of sound on the distribution of cod in quarter 4 was evident, even when the fish numbers in the quarter did not change significantly or remained constant during sound production.

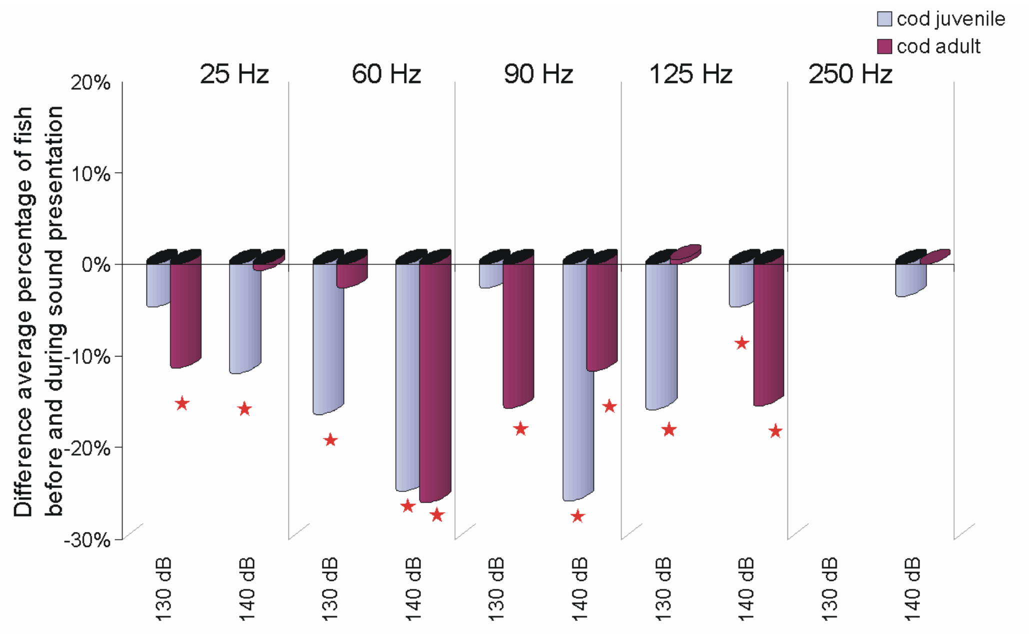


Fig. 49: Summary of the differences in the mean fish numbers [%] between the periods before and during sound production in cod. Significant differences (Mann-Whitney-U-Test (α = 5%) with Bonferroni correction) are marked with an asterisk.

4.2.2 Experiments using plaice

4.2.2.1 Observations without sound production

4.2.2.1.1 General behaviour

The predominant behaviour observed was resting on the tank bottom, which included small movements during resting behaviour and turning on the spot. On average, 75% of the juveniles and 86% of the adult plaice were resting at any time. Most resting fish were located near the water inflow in tank quarter 4 (Fig. 50) orientated into the inflow. Swimming behaviour was observed close to the tank bottom, near the water surface and in the water column. Occasionally plaice swam upright with their head out of the water or upside down near the bottom scraping their back along the bottom.

4.2.2.1.2 Distribution of plaice in the tank

Plaice showed a pronounced preference for quarter 4 with the average median of individuals being 65% for juvenile and 78% for adults.

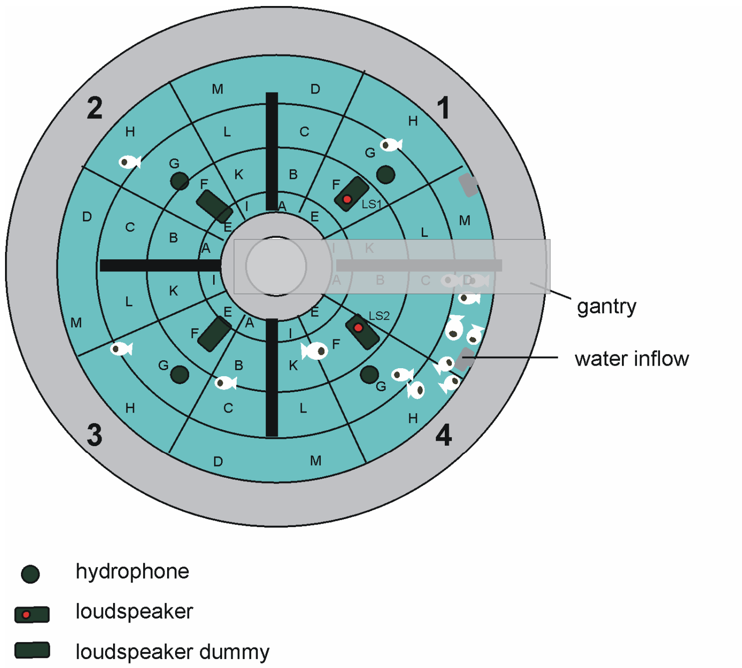


Fig. 50: Sketch of the tank showing the position of the sections A - M in every quarter of the tank and the area in quarter 4 that was preferred by plaice. The accumulation of fish symbols in quarter 4 indicate the preferred area while the other parts of the tank are less frequently used by the fish.

The distribution of plaice in quarter 4, summarized for all experiments, showed a preference for the section in which the water inflow was located (D) which was most pronounced in juveniles. 80% of juveniles and 51% of adults were located in the sections D, H and C (Fig. 51)

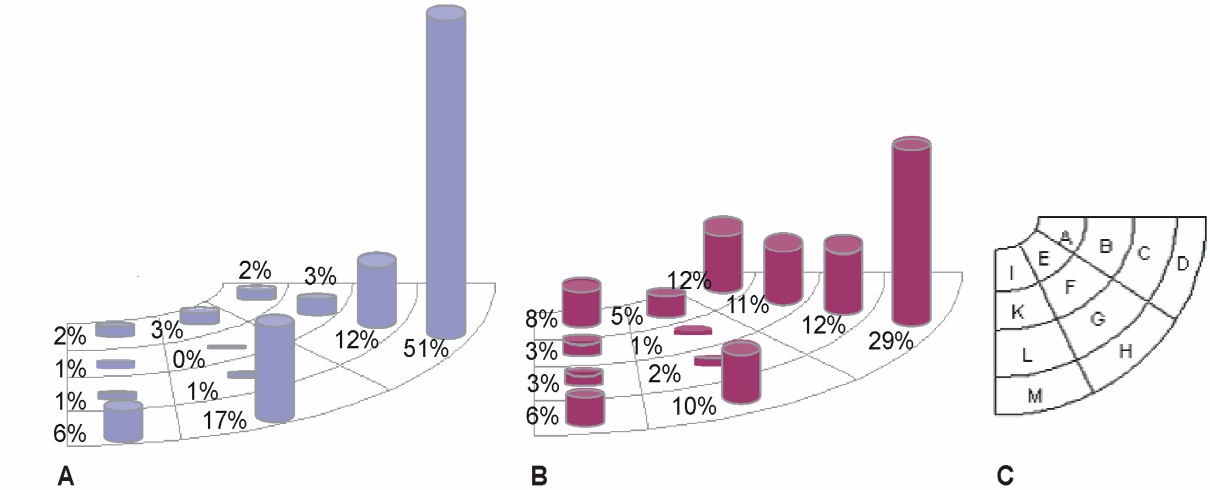


Fig. 51: Distribution of juvenile (**A**) and adult (**B**) plaice displayed as the percentage of fish present in quarter 4. Data from periods without sound production pooled for all experiments. **C**: Position of sections.

Whilst juvenile plaice showed a strong preference for the areas of highest current flow (section D) adult plaice showed a less clumped distribution also occupying areas along the barriers and tank walls with lower current flow. The lowest numbers were recorded in sections F and G, located in the middle of this part of the tank and not bounded by any barriers or walls.

4.2.2.1.3 Diel rhythm in plaice

Examination of the figures showing the number of resting and active fish present in quarter 4 counted every 15 minutes (appendix Fig. A 45 to Fig. A 63) revealed a diel pattern in plaice that was most pronounced in adult fish. Adult plaice were usually active from the morning until the light was switched off in the early evening. But it needs to be mentioned again that on average only 14% of adult plaice were active. During darkness (red light period) the number of resting fish increased to a peak in the middle of the night and decreased again in the early morning while the number of active fish was lowest in the middle of the night (Fig. 52). In juvenile plaice a tendency to higher numbers of resting fish at night could be seen but the number of active fish appeared to vary independently of the light regime (Fig. 53).

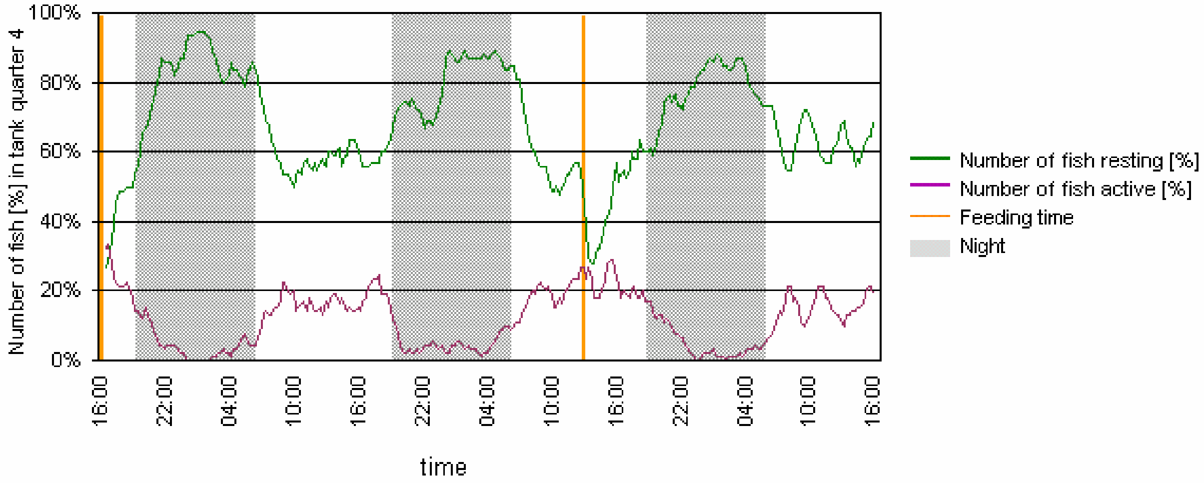


Fig. 52: Diel rhythm in adult plaice (experiment 125 Hz, 140 dB re 1µPa). Resting and active fish in quarter 4 as percentage of all fish in the tank (N = 18) shown for a period of 72 hours. Figure smoothed.

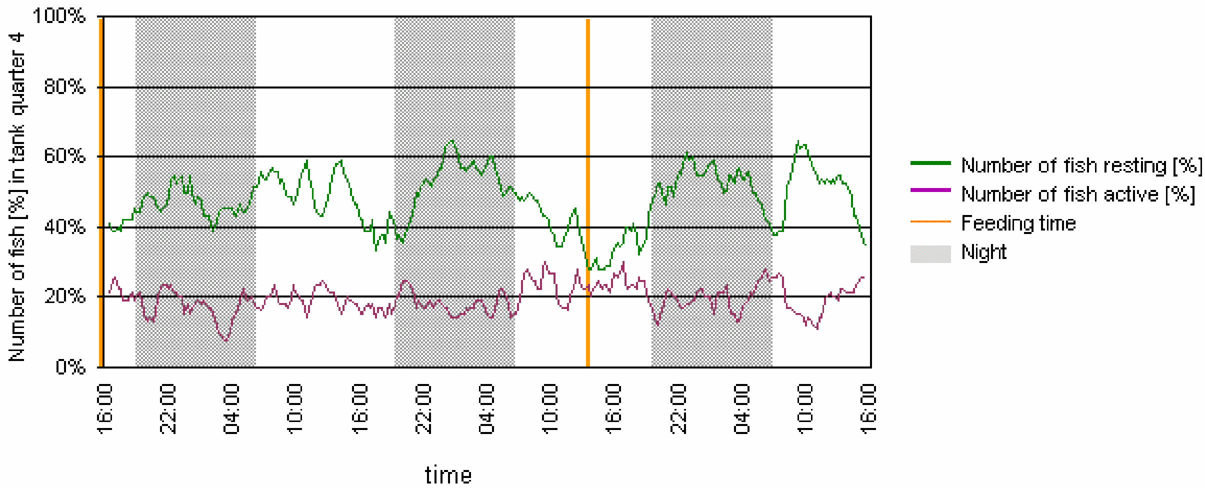


Fig. 53: Diel rhythm in juvenile plaice (experiment 125 Hz, 140 dB re 1µPa). Resting and active fish in quarter 4 as percentage of all fish in the tank (N = 18) shown for a period of 72 hours. Figure smoothed.

It is clear from Fig. 54 that the diel rhythm observed in adult plaice appeared only after a longer period of acclimatisation to the tank environment.

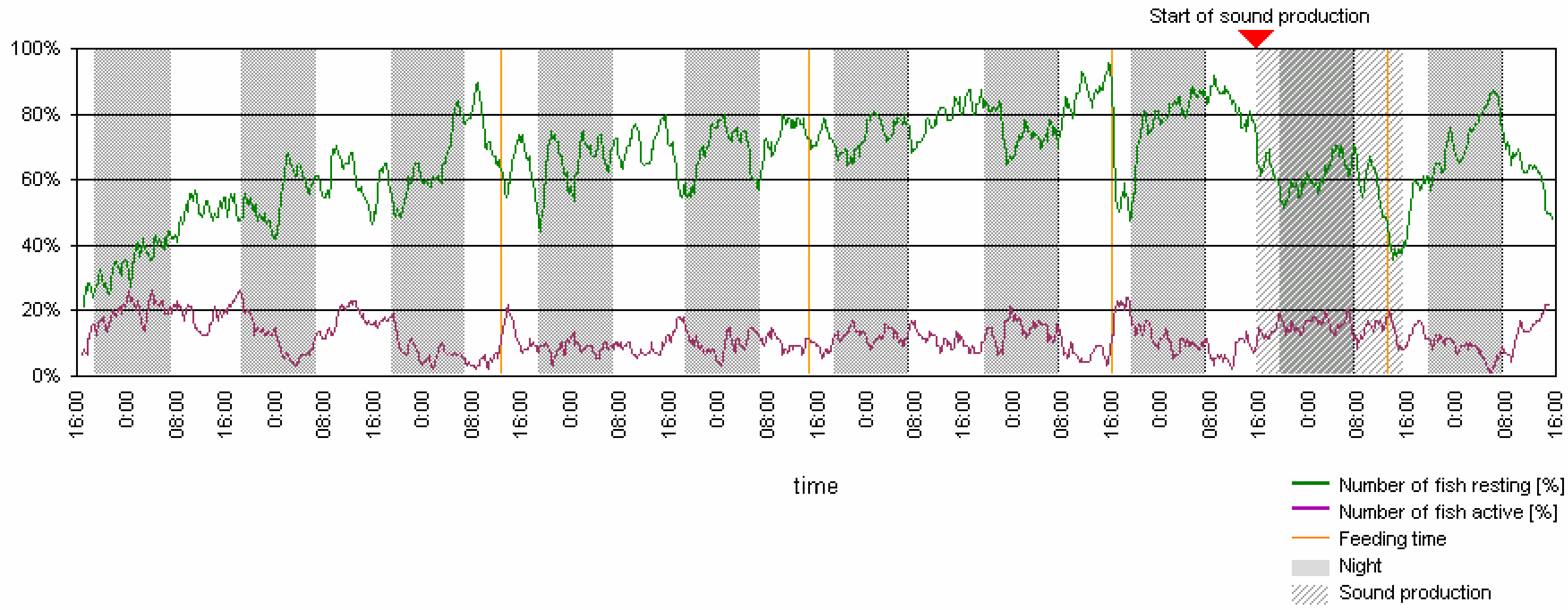


Fig. 54: Percentage of resting and active adult plaice in quarter 4 of all fish in the tank (N = 19) is shown for a period of 10 days after release of fish into the tank. The first sound experiment of 25 Hz at a sound level of 130 dB re 1µPa started at day 9. Figure smoothed.

Activity levels

Juvenile plaice showed a relatively constant level of about 15% of all fish active in quarter 4. Activity levels of adult plaice were lower with an overall average of about 11% but varying from 21% during the day and 5% during night periods. The activity pattern in each experimental series showed a high degree of similarity therefore the results were pooled to give an overall pattern of the diel activity pattern of juvenile and adult plaice in the experiments (Fig. 55). The figures show the average proportion (median) of active or resting fish in quarter 4 divided in day and night periods for a duration of 72 hours.

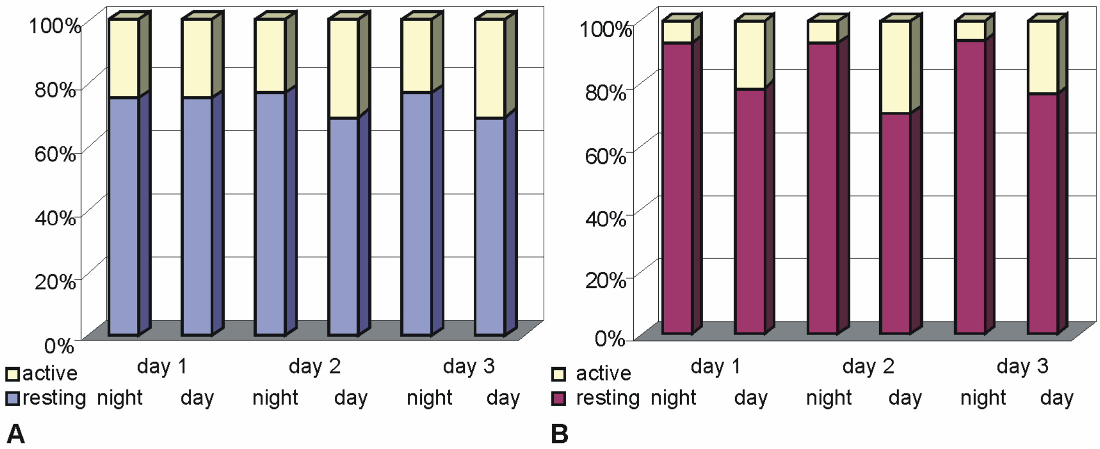


Fig. 55: Percentage of active and resting behaviour (median) of juvenile (**A**) and adult (**B**) plaice in quarter 4 during a 72 h observation period. The figure contains the pooled results of all experiments.

Presence in quarter 4 during day and night

The number of juvenile plaice present in quarter 4 did not show any clear diel pattern. In nine days the fish numbers present in quarter 4 were higher at night than in daytime. In 11 cases the number of fish was higher in daytime than at night and in six days the fish numbers in quarter 4 were about the same at day and night. As an example Fig. 56 gives the numbers of juvenile plaice present in this quarter during two 24 hour periods divided in day and night showing lower fish numbers at night in the first 24 hour period and higher numbers at night during the second 24h period.

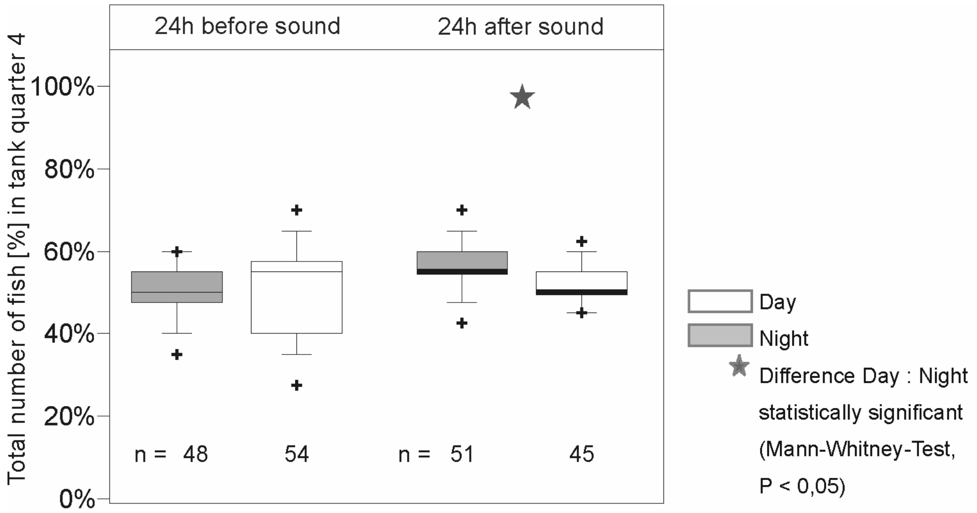


Fig. 56: Percentage of juvenile plaice in quarter 4 of all fish in the tank at day and night shown for two days as an example. Significant differences between day and night are marked by an asterisk above the Box-Whisker-Plots. n = number of pictures evaluated.

In contrast, adult plaice showed a diel pattern with higher fish numbers in quarter 4 at night compared with the daylight period in 28 out of 30 evaluated 24h periods An example is given in Fig. 57. During the remaining two 24h periods the average fish numbers present during the day and night was on about the same level. Statistical results are summarized in Table A 7 (appendix).

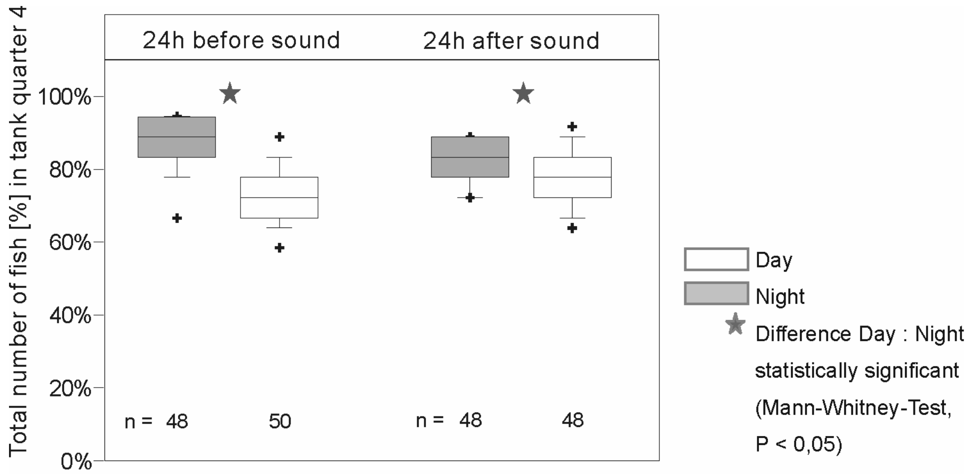


Fig. 57: Percentage of adult plaice at day and night in quarter 4 of all fish in the tank shown for two days as an example. Significant differences between day and night are marked by an asterisk above the Box-Whisker-Plots. n = number of pictures evaluated.

The lower number of adult plaice present in quarter 4 during the day was due to the higher activity levels during daylight hours. While fish rested mainly in quarter 4, when active, they moved around the whole tank and therefore the numbers present in quarter 4 were reduced.

4.2.2.1.4 Feeding

The fish were fed every second day during the experiments and the food was distributed evenly over the quarters of the tank. This led to movements of the fish leaving quarter 4 to search for food in other parts of the tank which became obvious in many of the figures (Fig. A 45a to Fig. A 63a, appendix). Although fish located the food relatively quickly, it could take up to three hours before numbers returned to pre-feeding levels. Two examples of the influence of feeding on the distribution of fish in the tank are given in Fig. 58A and B.

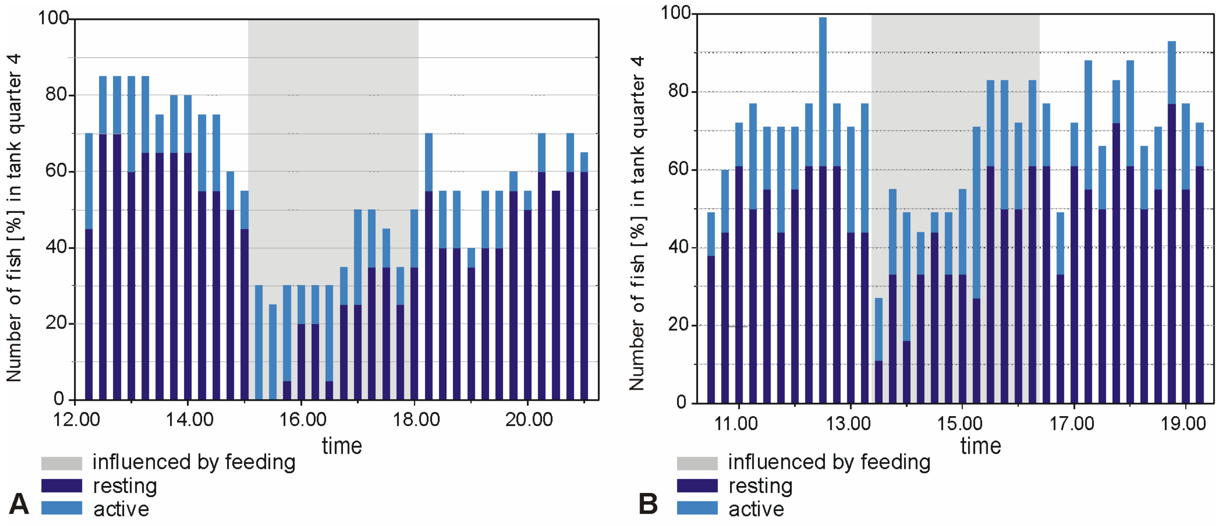


Fig. 58: Number and behaviour of juvenile (**A**) and adult (**B**) plaice in quarter 4 three hours before and six hours after a feeding. The number of fish in quarter 4 decreased after feeding when the fish moved to other parts of the tank in search for food and slowly returned to the preferred quarter after a period of up to three hours (coloured grey).

These changes in distribution were found to have a small, statistically non-significant influence on the analysis and therefore these periods were not excluded from data analysis.

4.2.2.2 Behaviour of plaice during sound production

The results of the behavioural experiments using plaice are displayed as box plots showing the fish numbers present in quarter 4 in the 24 hour periods before, during and after sound production. The sound was presented in quarter 4. All experiments using the same frequency are displayed and described together (Fig. 59 to Fig. 63). Statistically significant differences between periods are marked with asterisks. Additionally the average fish numbers (median) present in the periods before, during and after sound and the p-values of the Mann-Whitney-Test with Bonferroni-correction for significant differences between the different periods are given in Table 13 to Table 17. Additional figures showing the overall fish numbers present in quarter 4, the fish numbers divided into active and resting and box plots showing the fish numbers in the periods before, during and after sound during day and night as well as divided into active and resting can be found in Fig. A 45 to Fig. A 63 (appendix). All statistical results of the Kruskal-Wallis-test and the Mann-Whitney-Test are summarized in Table A 8 (appendix).

4.2.2.2.1 Tested frequency of 25 Hz

In the experiment using juvenile plaice during sound production of 25 Hz at 130 dB re 1µPa lower fish numbers were observed in quarter 4 before sound production compared with the numbers during and after sound. The differences were non-significant. Due to data loss of the first 12 hours of sound production the box-plot during sound production contains mostly data of the daylight period.

In the experiments using adult plaice significant differences appeared only during sound production at 130 dB re 1µPa. The number of fish in the period before sound was significantly higher than in the during and after sound periods. While the sound of 140 dB re 1µPa was turned on, the average fish numbers decreased but the differences were non-significant.

Table 13: Average fish numbers (median) present in quarter 4 in experiments using plaice during sound production of 25 Hz

Juvenile plaice	Before sound	During sound	After sound
25 Hz, 130 dB re 1µPa	50.00%	55.00%	55.00%
Adult plaice	**Before sound**	**During sound**	**After sound**
25 Hz, 130 dB re 1µPa	89.47% (p = 0.0000 before–during; p = 0.0001 before–after)	73.68%	78.95%
25 Hz, 140 dB re 1µPa	72.22%	66.67%	77.78%

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

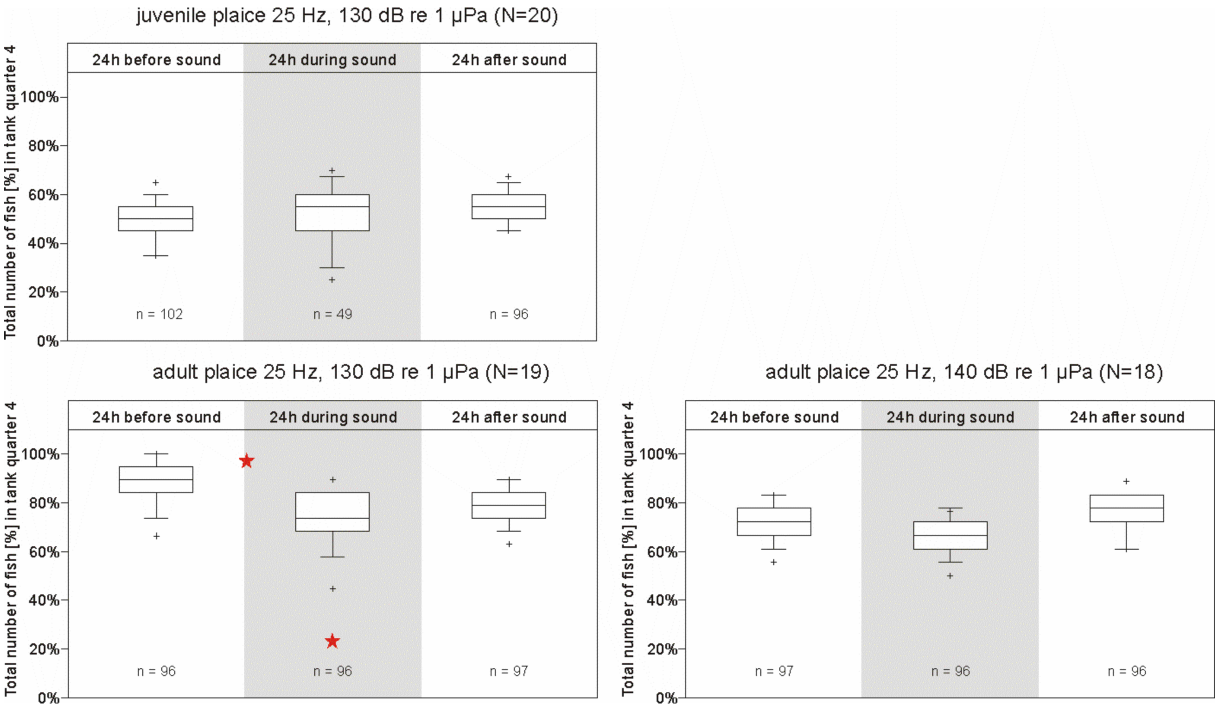


Fig. 59: Proportion of juvenile and adult plaice (median) present in quarter 4 before, during and after sound production of 25 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box

4.2.2.2.2 Tested frequency of 60 Hz

The production of 60 Hz led to ambiguous results. The number of juvenile fish in quarter 4 decreased during sound production at 130 dB re 1µPa but only the difference between the periods before and after sound was statistically significant. In the experiment at 140 dB re 1µPa the fish numbers increased significantly during sound production and returned to the pre-noise level after the sound was switched off.

In both experiments using adult plaice the fish numbers increased in quarter 4 during sound production and decreased again after the sound was switched off but the differences were not statistically significant.

Table 14: Average fish numbers (median) present in quarter 4 in experiments using plaice during sound production of 60 Hz

Juvenile plaice	Before sound	During sound	After sound
60 Hz, 130 dB re 1µPa	70.00%	60.00%	60.00%
	p = 0.0150 (before vs. after)		
60 Hz, 140 dB re 1µPa	60.00%	75.00%	60.00%
	p = 0.0004 (before vs. during)	p = 0.0012 (during vs. after)	
Adult plaice	**Before sound**	**During sound**	**After sound**
60 Hz, 130 dB re 1µPa	77.78%	88.89%	83.33%
60 Hz, 140 dB re 1µPa	78.95%	84.21%	78.95%

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

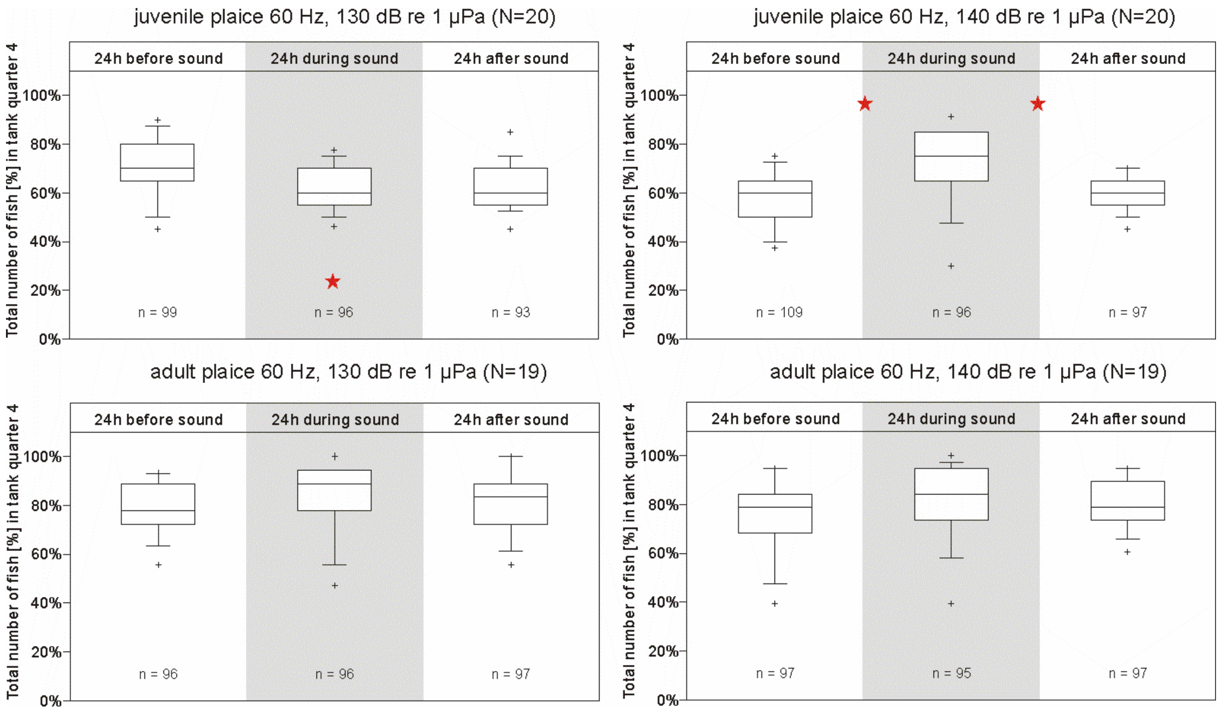


Fig. 60: Proportion of juvenile and adult plaice (median) present in quarter 4 before, during and after sound production of 60 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

4.2.2.2.3 Tested frequency of 90 Hz

In all experiments using a sound frequency of 90 Hz the fish numbers in quarter 4 decreased while the sound was turned on, although results were only significant for juvenile plaice at 140 dB re 1µPa.

Table 15: Average fish numbers (median) present in quarter 4 in experiments using plaice during sound production of 90 Hz

Juvenile plaice	Before sound	During sound	After sound
90 Hz, 130 dB re 1µPa	65.00%	65.00%	75.00%
90 Hz, 140 dB re 1µPa	76.47%	64.71%	64.71%
	p = 0.0023 (before vs. during); p = 0.0029 (before vs. after)		
Adult plaice	**Before sound**	**During sound**	**After sound**
90 Hz, 130 dB re 1µPa	78.95%	73.68%	78.95%
90 Hz, 140 dB re 1µPa	72.22%	66.67%	72.22%

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

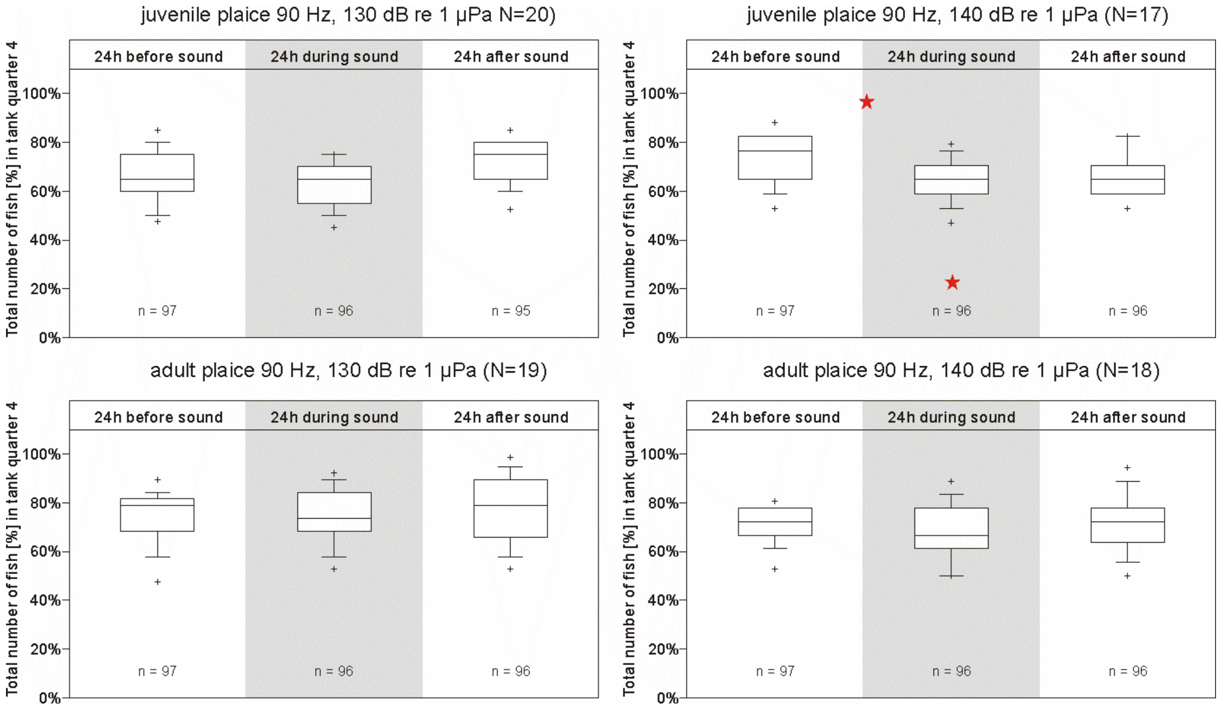


Fig. 61: Proportion of juvenile and adult plaice (median) present in quarter 4 before, during and after sound production of 90 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

4.2.2.2.4 Tested frequency of 125 Hz

During sound production of 125 Hz the differences between periods with and without sound were relatively small in both fish groups. Significant differences appeared only during sound production at 130 dB re 1µPa with increasing numbers of juvenile plaice after the sound was switched off.

Table 16: Average fish numbers (median) present in quarter 4 in experiments using plaice during sound production of 125 Hz

Juvenile plaice	Before sound	During sound	After sound
125 Hz, 130 dB re 1µPa	57.50%	55.00%	65.00%
		p = 0.0020 (During vs. After); p = 0.0001 (Before vs. After)	
125 Hz, 140 dB re 1µPa	66.67%	66.67%	72.22%
Adult plaice	**Before sound**	**During sound**	**After sound**
125 Hz, 130 dB re 1µPa	68.42%	68.42%	73.68%
125 Hz, 140 dB re 1µPa	83.33%	77.78%	83.33%

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

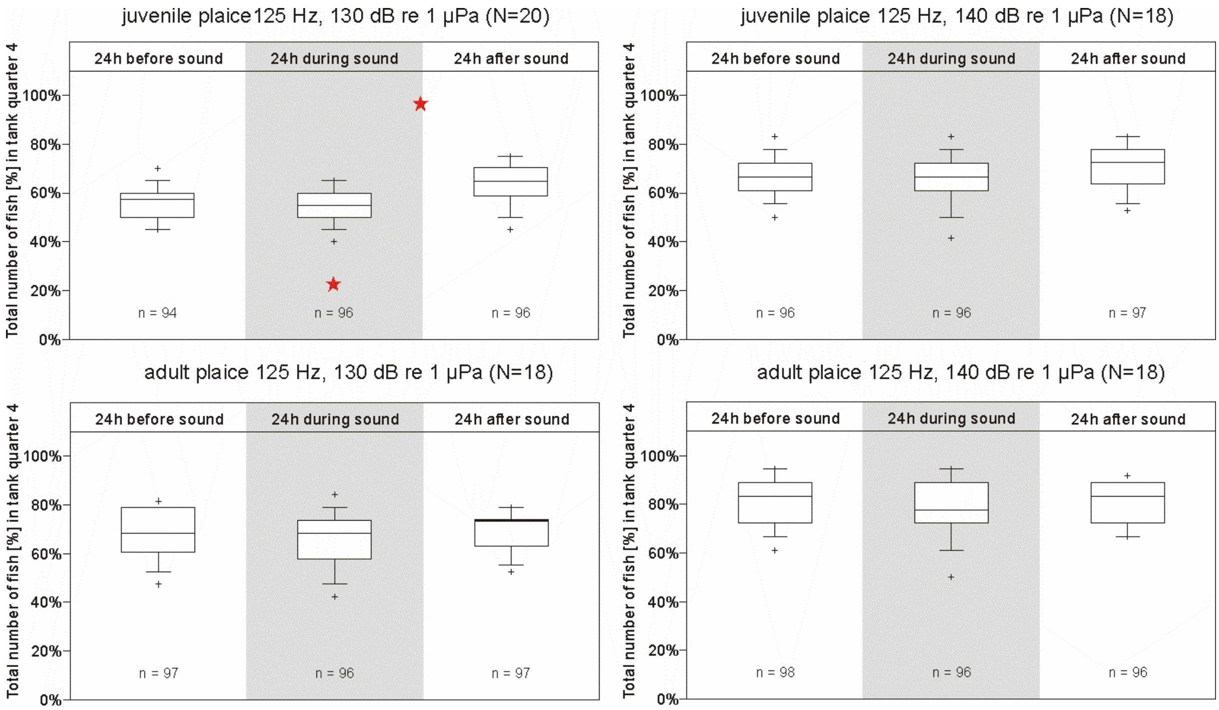


Fig. 62: Proportion of juvenile and adult plaice (median) present in quarter 4 before, during and after sound production of 125 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

4.2.2.2.5 Tested frequency of 250 Hz

The results of the highest tested frequency of 250 Hz were ambiguous both with decreasing and increasing fish numbers during sound production. Significant differences only appeared in both experiments using juvenile plaice where the fish numbers increased in the after sound period. In the experiments using adult plaice slightly higher fish numbers were observed in quarter 4 during sound production in both age groups.

Table 17: Average fish numbers (median) present in quarter 4 in experiments using plaice during sound production of 250 Hz.

Juvenile plaice	Before sound	During sound	After sound
		p = 0.0001	
250 Hz, 130 dB re 1µPa	63.16%	57.89%	73.68%
		p = 0.0006	
		p = 0.0055	
250 Hz, 140 dB re 1µPa	55.00%	60.00%	60.00%
Adult plaice	**Before sound**	**During sound**	**After sound**
250 Hz, 130 dB re 1µPa	72.22%	83.33%	77.78%
250 Hz, 140 dB re 1µPa	78.95%	84.21%	84.21%

P-values of the Mann-Whitney-Test with Bonferroni-correction are given for significant differences between periods before, during and after sound production.

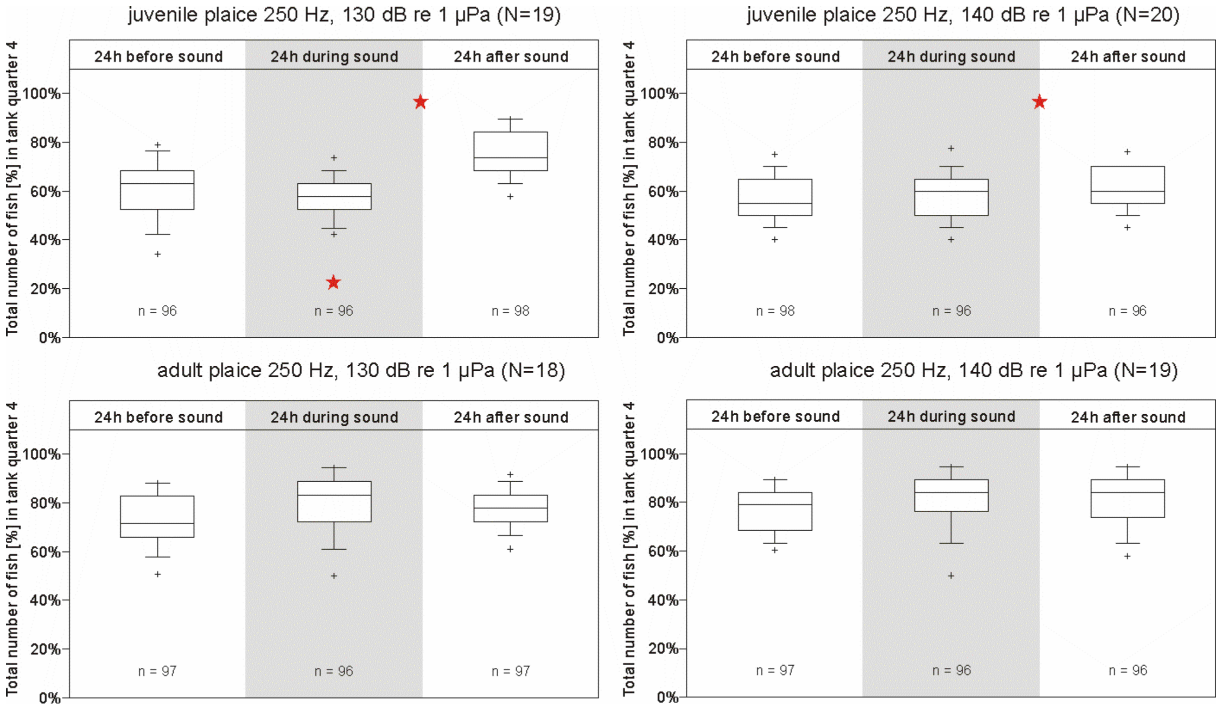


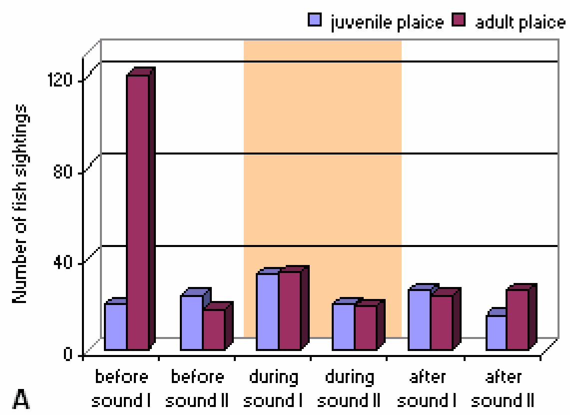
Fig. 63: Proportion of juvenile and adult plaice (median) present in quarter 4 before, during and after sound production of 250 Hz at sound levels of 130 dB re 1µPa (left) and 140 dB re 1µPa (right). N = fish numbers present in the tank, n = number of pictures evaluated. Significant differences between 24 hours periods are marked with asterisks (before-during and during-after sound above box plots, before-after sound below box plots).

4.2.2.2.6 Loudspeaker vicinity evaluation

As a further analysis of the data, changes in the fish numbers in the direct vicinity of the sound source during sound production were assessed. At the bottom of the tank, where most of the plaice were present the sound pressure difference in quarter 4 was between 17 and nearly 38 dB depending on frequency and sound level. In the 1 m vicinity of the loudspeaker the sound levels decreased by only 5 to 19 dB and fish would be exposed to far higher sound levels in this area than in other parts of quarter 4.

Three of the sound experiments using plaice were evaluated in detail. In two of these experiments significant lower fish numbers in the whole of quarter 4 were observed during sound production. In the third, the fish numbers in the whole of quarter 4 did not change significantly.

The results of the 1 m range analysis in two of the three experiments showed widely varying numbers of plaice entering the circle around the loudspeaker independent of sound production (Fig. 64). When the duration of presence of every fish entering the vicinity of the loudspeaker was analysed, any pattern related to the sound did not appear but a tendency for longer stays was observed in adult plaice (Fig. 66, Fig. 65).



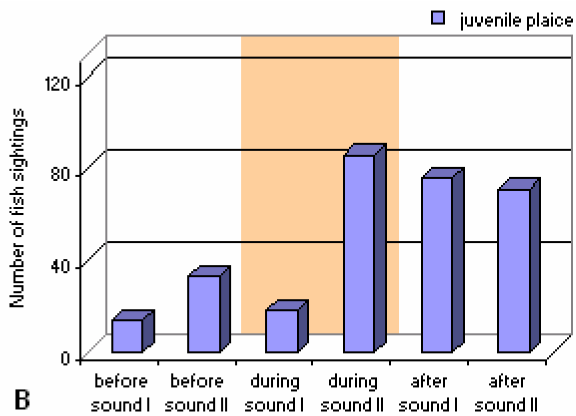


Fig. 64: Number of sightings of plaice entering the area of one meter around the loudspeaker in 30 minute periods at the beginning and the end of the phases before, during and after sound production each. The sound period is coloured. **A**: During the experiments (juvenile 60 Hz, 140 dB, adult 25 Hz, 130 dB) the fish numbers in the whole of quarter 4 decreased significantly. **B**: The experiment showed no significant changes in juvenile plaice numbers in the whole of quarter 4 during sound production 250 Hz at 140 dB.

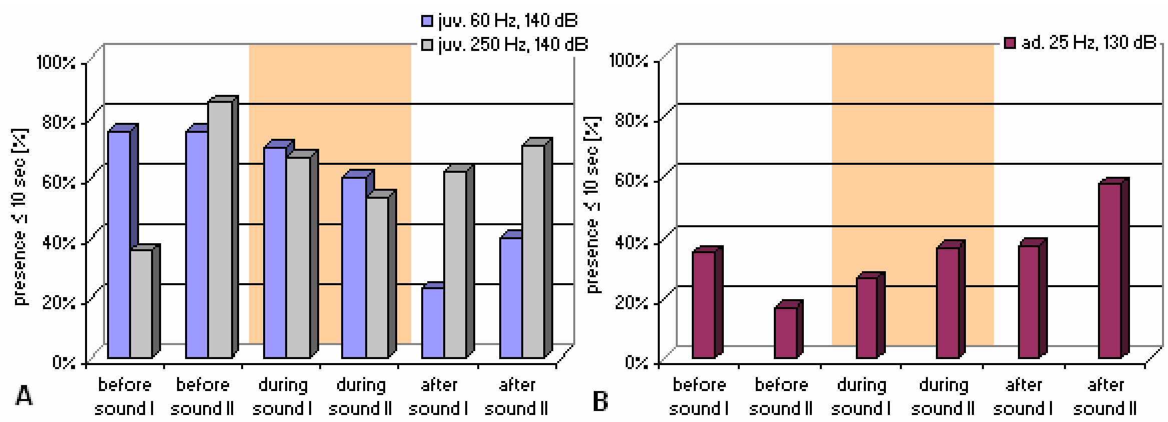


Fig. 65: Percentage of short duration presence of 10 or less seconds in a circle of one meter around the loudspeaker in 30 minute periods at the beginning and the end of the phases before, during and after sound production each. The sound period is coloured. **A**: Experiments using juvenile plaice. **B**: Experiment using adult plaice.

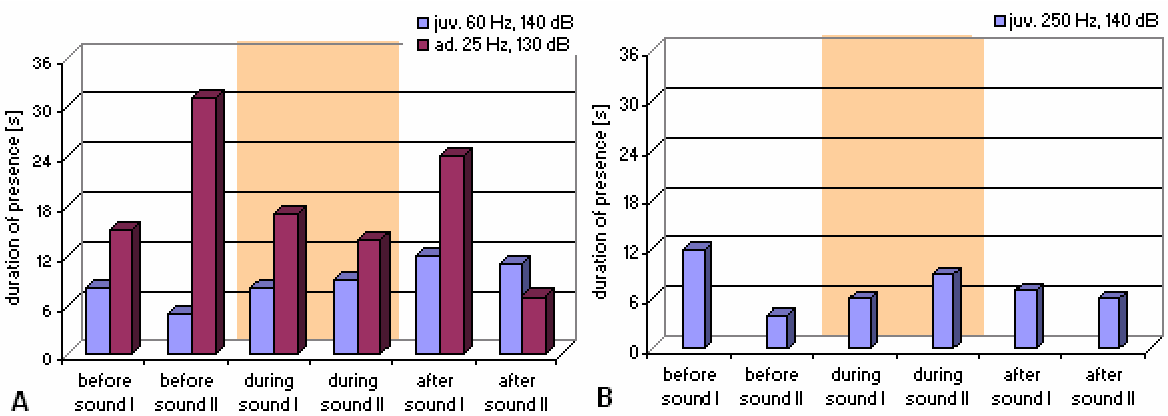


Fig. 66: Average duration of presence (median) in a circle of one meter around the loudspeaker in 30 minute periods at the beginning and the end of the phases before, during and after sound production each. The sound period is coloured. **A**: Experiments with and **B** without significant lower numbers of plaice in quarter 4 during sound production in the overall evaluation.

4.2.2.2.7 Habituation of plaice to sound

Generally fish were given a period of seven days to adapt to the tank environment before the sound experiments started. During the first day most fish settled in quarter 4. To investigate the effect of sound on the settlement behaviour six juvenile plaice (TL 24 to 30 cm) were introduced into the tank with sound (60 Hz, 140 dB re 1µPa) already turned on. The sound was switched off after 45 hours and the distribution of fish evaluated for another 14 hours. The results of this experiment were compared with the adaptation periods of the previous experiments (Fig. 67).

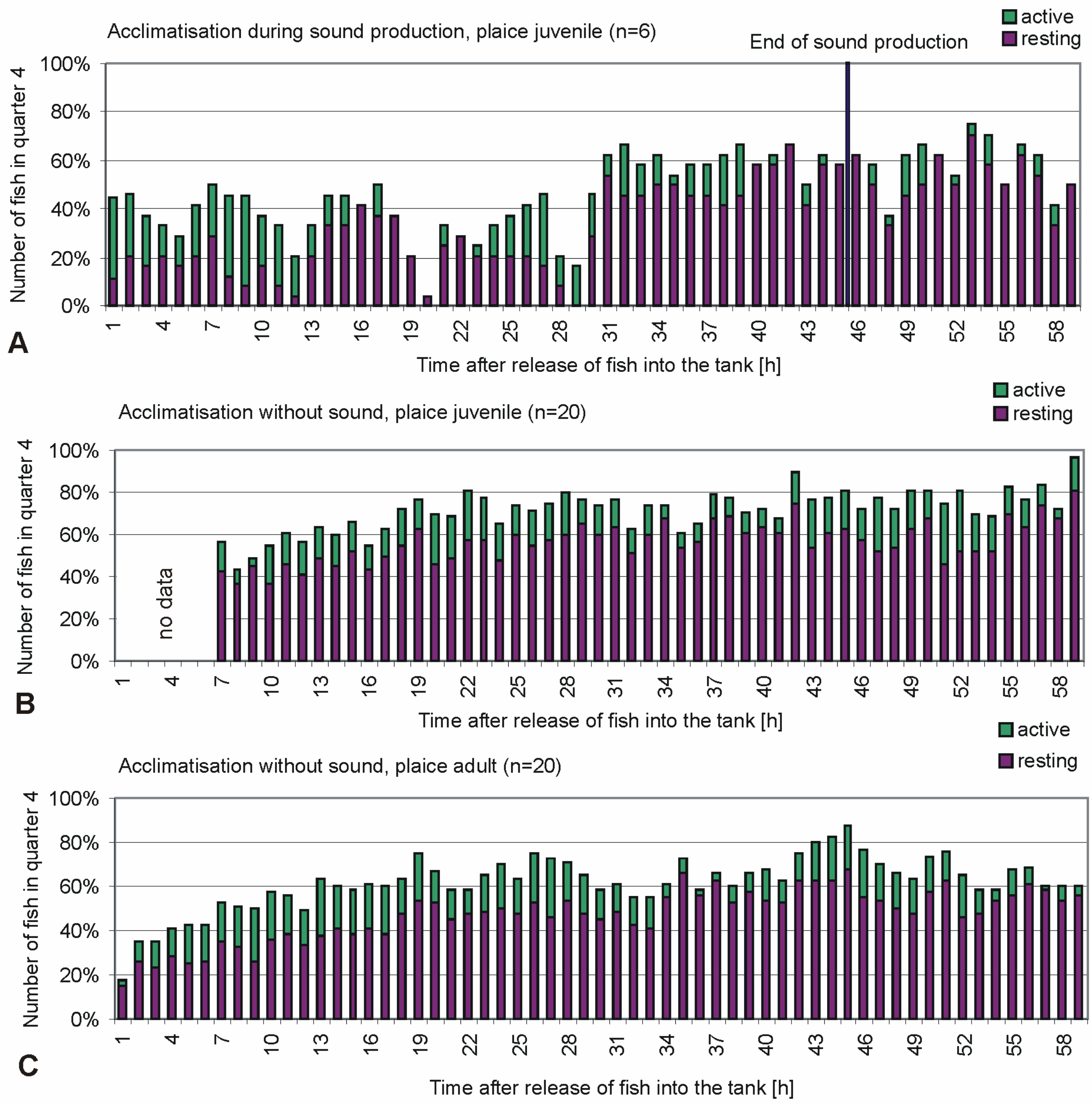


Fig. 67: Settlement of plaice in tank quarter 4. The hourly percentage of fish in quarter 4 is displayed for three experiments **A**: Juvenile plaice released into the tank during sound production (60 Hz 140 dB re 1µPa) **B**: Juvenile plaice released into the tank without sound **C**: Adult plaice released into the tank without sound.

Over a period of 24 hours starting six hours after release of fish into the tank, an average 65% of the juvenile and 60% (medians) of the adult plaice settled in tank quarter 4. During the same period of time only about a third of the fish settled in quarter 4 when sound was being produced. Only after about 30 hours did the proportion of fish in quarter 4 reach similar levels to the acclimatization periods without sound. Numbers remained relatively constant over the next 30 hour period.

The results demonstrate a delayed settlement of juvenile plaice in quarter 4 during sound production compared with settlement without sound but indicated habituation to the sound after a period of about 30 hours.

In the habituation with sound experiment about two third of the juvenile plaice were present in the sections M, H and K (Fig. 68D) in a 24 hours period starting 6 hours after release of the fish into the tank (Fig. 68A). These sections are closest to the only entrance into the quarter. The sound source was located in section F. In section D, preferred by juvenile and adult plaice during acclimation without sound production, only 11% of the fish were present during this period. Looking at the period until the end of the sound production (31^{st} to the 45^{th} hour) the distribution of fish in quarter 4 did not change a lot (Fig. 68B). Most of the fish were present in the sections near the entrance. The highest fish numbers were present in sections I and M containing 29% and 31% of the fish. Section D contained less than 7%, and section B (close to the loudspeaker) contained 11%. After the sound was switched off the distribution of fish in quarter 4 shifted closer to the loudspeaker (Fig. 68C) with about 40% of the fish present in the sections A, B and E. The fish numbers present in section D were still very low, at less than 4% and fish numbers present in section I remained on about the same level of 29% as in the period before turning the sound off.

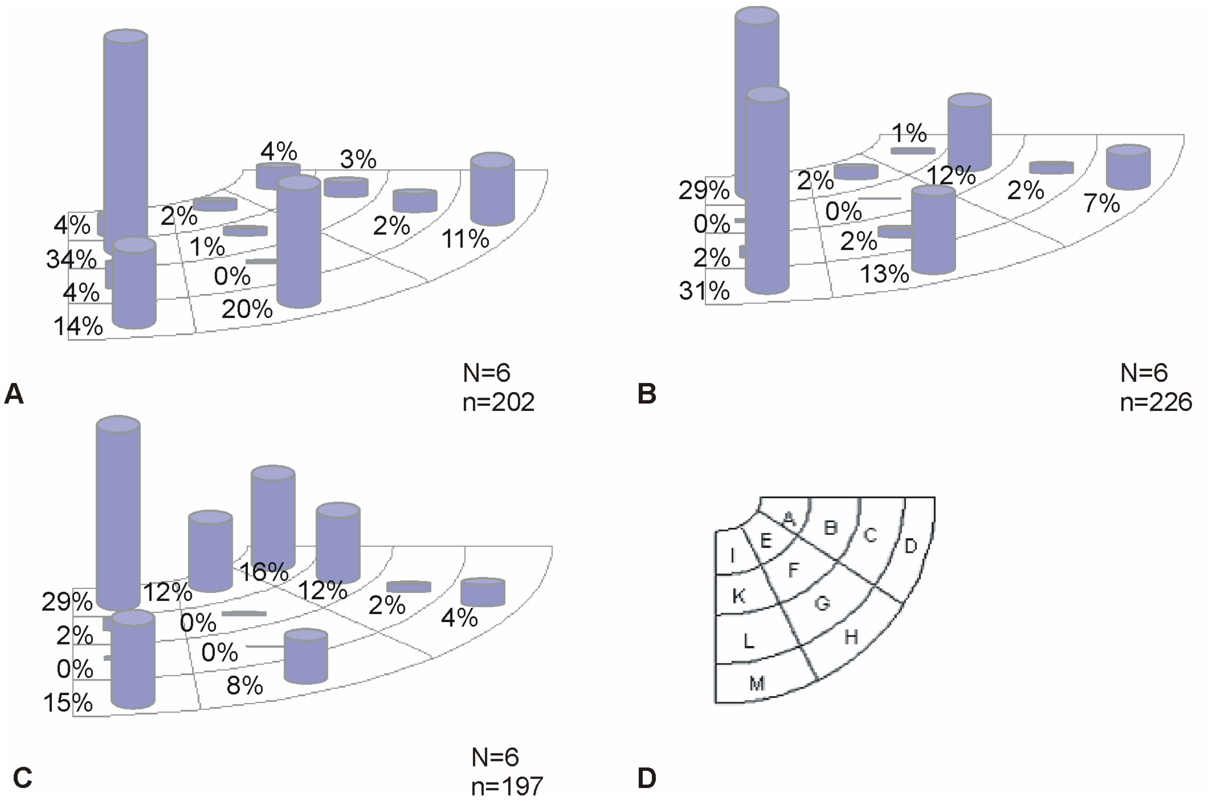


Fig. 68: Percentage of juvenile plaice in the sections of quarter 4 during a 59 hours acclimatisation period with sound production of 60 Hz at 140 dB re 1µPa. **A**: 24 hours period starting 6 hours after release of fish into the tank. **B**: 15 hours period (31^{st} to 45^{th} hour) before turning the sound off. **C**: Period of 14 hours after turning the sound off. **D**: Position of sections in quarter 4. N = Fish numbers in the tank, n = number of fish sightings.

Striking were differences in distribution of juvenile and adult plaice in quarter 4 compared with the habituation of juvenile plaice during sound production. During acclimatisation of juvenile plaice (Fig. 69A) half of the fish were present in section D and another 10% to 11% each were found in the sections M, H and C. In adult plaice most of the fish were present in the sections next to the outer tank wall. Sections D and H contained 30% and 27% of the fish, in section M more than 13% of the fish were present (Fig. 69B).

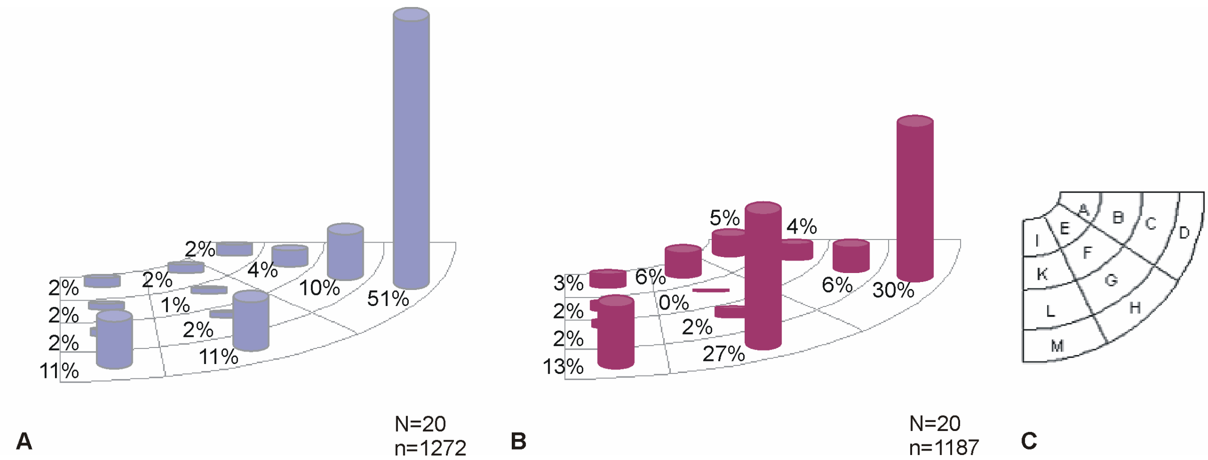


Fig. 69: Percentage of juvenile (**A**) and adult (**B**) plaice in the sections of quarter 4 during a 24 hours acclimatisation period starting 6 hours after release of fish into the tank. No sound had been switched on. N = Fish numbers in the tank, n = numbers of fish sightings.

Examining the sound level distribution in quarter 4 (Fig. 70), the highest sound levels appeared in the sections A, B, F and E while the lowest sound levels were measured in the sections L, M and H. The relatively high amount of fish near the entrance area during sound production and the shift to the sections A, B and E after the sound was switched off indicate avoidance behaviour of juvenile plaice to the sound. On the other hand the measured sound levels of 143 to 148 dB re 1µPa in section I which contained about 29% of the fish were relatively high. This suggests that the sound level may not be not the only aspect that influenced the distribution of fish.

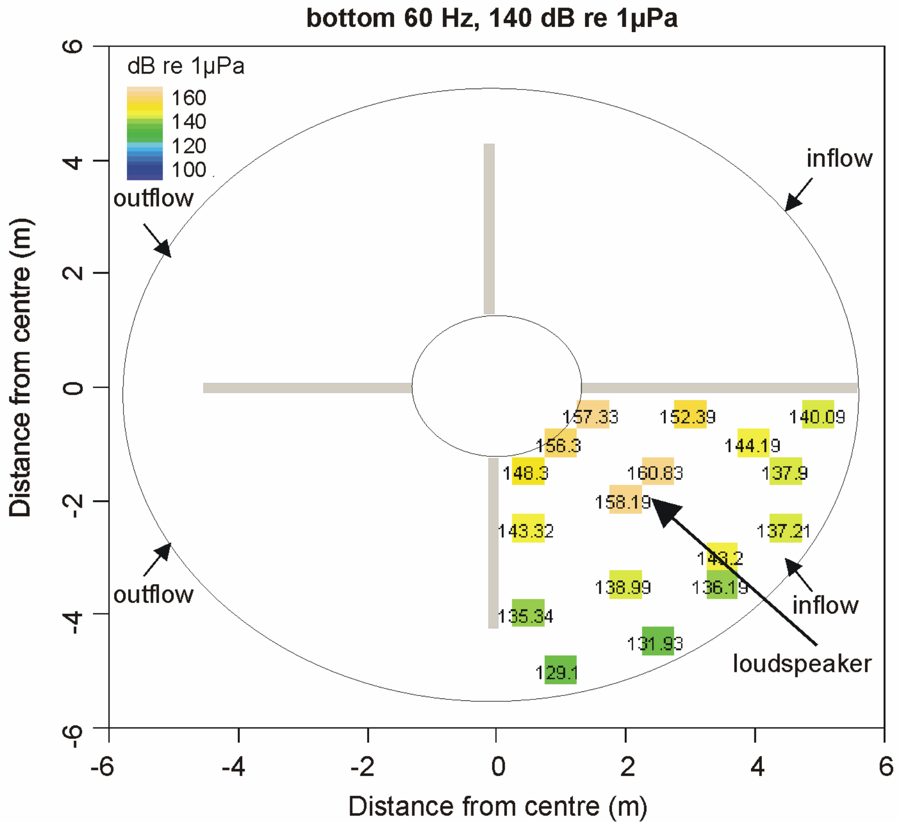


Fig. 70: Sound field near the bottom in quarter 4 during sound production of 60 Hz at 140 dB re 1µPa.

4.2.2.3 Recapitulation of plaice results

The results of the sound experiments suggest possible avoidance and attraction of plaice to sound. However, a clear pattern could not be seen. To summarize the results of plaice the differences in the arithmetic mean of the fish numbers present in quarter 4 at the periods before and during sound production are displayed in Fig. 71. Significantly lower fish numbers could be observed in one experiment each at the frequencies 25 and 90 Hz, while the number of fish in quarter 4 increased significantly in one experiment using 60 Hz. If the day and night periods are separated, the pattern becomes even less clear with different reactions during day and night of the same period (Fig. 72). A comparison of the periods before, during and after sound summarized for all experiments using the same group of fish did not show significant differences (Friedman-Test) between the periods with and without sound production.

Similarly in the loudspeaker vicinity no reaction of plaice to sound was found. However the settlement of juvenile plaice in the preferred area of the tank seemed to be delayed by sound.

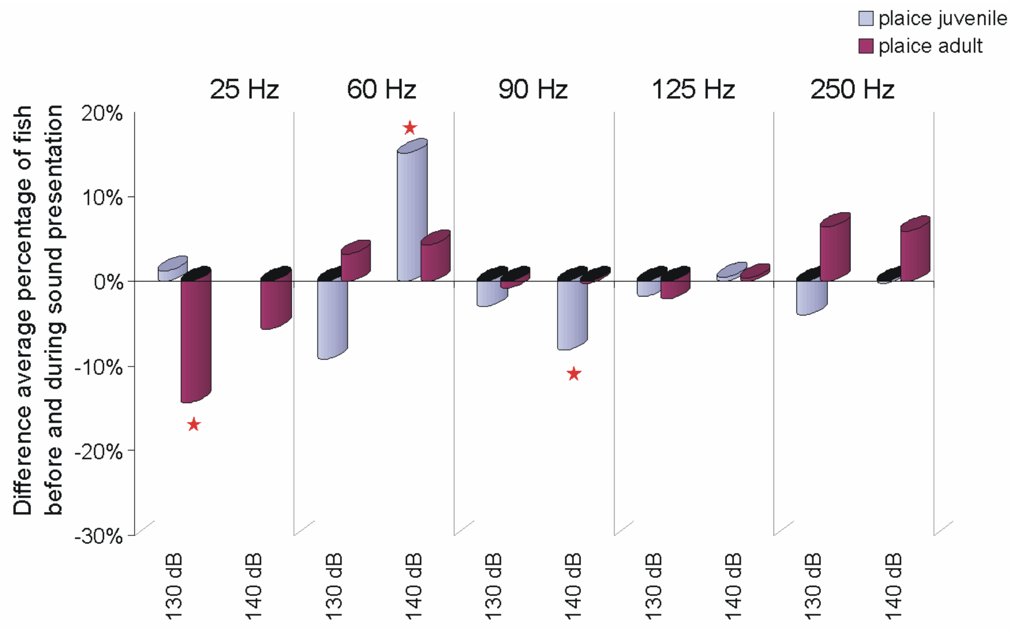


Fig. 71: Summary of the differences in the mean fish numbers [%] between the periods before and during sound production in plaice. Significant differences (Mann-Whitney-U-Test (α = 5%) with Bonferroni correction) are marked with an asterisk. Negative values indicate decreasing fish numbers while positive numbers show an increase of fish in quarter 4 during sound.

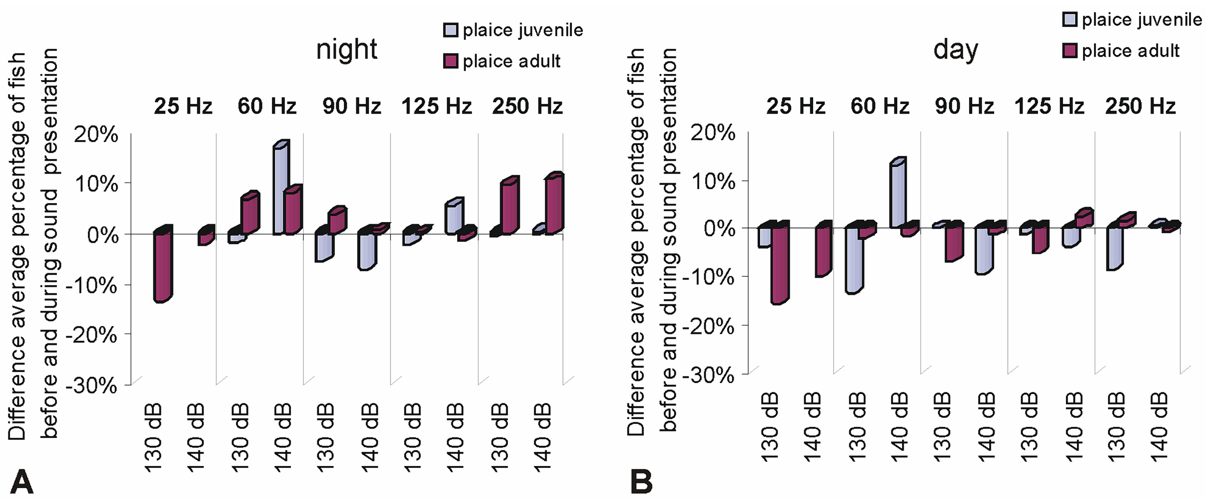


Fig. 72: Summary of the differences in the mean plaice numbers [%] between the periods before and during sound production at night (**A**) and day (**B**). Negative values indicate decreasing fish numbers while positive numbers show an increase of fish in quarter 4 during sound.

5 Discussion

5.1 Experimental results

5.1.1 Hydroacoustic field in the tank

The most important challenge in carrying out these experiments was the creation of a high sound pressure difference to expose the experimental fish to a complex sound field in the tank. In the direct vicinity of the sound source the sound reached a level comparable with the noise produced by a heavy truck that passes by at a speed of 64 km/h at a distance of 15 m (GREEN 1995) but in large parts of the tank the sound level corresponded with the noise in a quiet residential area at night. Therefore, the first question raised at the beginning of the project, that is whether the acoustic field in the tank could be controlled and the sound pressure difference could be increased can clearly be answered positively.

In all produced sound fields the average sound pressure levels in the quarters 2-4 exceeded the average background noise level. This indicates that masking of the produced sound by background noise did not limit the sound level decrease. Instead it seemed that after a quick decrease of sound level in the vicinity of the sound source the sound level decrease proceeded much slower and levelled out. This appeared at all frequencies on slightly different sound levels and differences in sound level became difficult to tell apart.

5.1.1.1 Effect of boundaries on the sound field

The acoustic field in a confined tank is complex compared with the sound field in an open space without limiting boundaries. This is particularly true in water where the wavelength of a given frequency is much longer than it is in air. Especially at the low frequencies to which fishes are sensitive, the wavelengths mainly go beyond the dimension of tanks. In the experiments carried out, the wavelengths of four out of five frequencies exceeded the ten meter diameter of the tank.

The use of Styrofoam barriers as sound boundaries increased the sound pressure differences in the tank. The high content of air in Styrofoam forms a water-air-interface, that is a soft boundary where most of the sound is reflected due to the different impedance of water and air (HAWKINS & MYRBERG 1983). But the sound pressure level only decreases to zero if the medium behind the soft boundary is a perfect pressure release medium (FREYTAG 1967). Therefore the thin Styrofoam sheets glued to marine plywood did not form perfect soft boundaries. The sound reducing effect of the Styrofoam barriers was strongest at 250 Hz but could also be seen at 125 Hz and 90 Hz.

FREYTAG (1967) described an exponential decrease of sound pressure level from the sound source towards pressure release boundaries. This effect could be observed towards the water surface in the tank.

5.1.1.2 Resonant frequency of the tank

FREYTAG (1967) calculated a resonant frequency for rectangular concrete tanks beneath which sound propagation is noticeably reduced. Using FREYTAG's formula at the annular radius of the tank of 3.68 m

$$\frac{\lambda}{2} = 3{,}68$$

a wavelength of 7.36 m was calculated, which equals a frequency of 195 Hz as resonant frequency of the tank. The reduced propagation of sound below the resonant frequency of the tank could explain the rapid sound level decrease in the vicinity of the sound source that was much higher than it would have been under far field conditions in open water.

Although the results of the lower frequencies differed from the results of the frequency of 250 Hz, the differences were not obvious enough to proof the existence of a resonance frequency of 195 Hz in the tank. It is likely that FREYTAG's formula (1967) cannot easily be transferred from rectangular to annular tanks. Whether the annular experimental tank with its thick concrete walls has a resonance frequency and in which frequency range it might be could not be determined.

5.1.1.3 Comparison of sound level decrease in the tank with field conditions

The sound level decrease in the tank greatly exceeded the transmission loss (TL) expected for shallow water conditions in the open sea, which can be calculated as TL = -10log(r) (r = distance [m]) (BASS & CLARK 2003). Therefore in shallow waters the transmission loss of sound over a distance of 2.2 m (distance from the sound source to the tank wall) would be about -3.5 dB. In the experimental tank divided by barriers the transmission loss at the same distance was 15 to 35 dB depending on frequency. To reach transmission loss of 15 to 35 dB as observed in the tank a distance of approximately 32 m to 3200 m would be necessary in shallow waters, while it would be approximately 6 m to 60 m in a free field in the open sea.

5.1.1.4 Hydrodynamic field

For many fish species, including plaice the relevant stimuli for sound detection are the hydrodynamic components (SAND & KARLSEN 1986, KARLSEN 1992b, ENGER et al. 1993). Unfortunately the attempt to measure particle displacement failed.

Theoretically the particle displacement in quarter 4 should be higher than in the other parts of the tank which showed lower sound pressure levels. The vertical particle displacement should decrease close to the tank bottom since the concrete acts as a rigid sound boundary (FREYTAG 1967). Therefore the particle movement at the bottom of the tank was likely to be lower than in the water column. This might be different from the sediment bottom prevalent in North and Baltic Sea, which forms a soft boundary (FREYTAG 1967).

5.1.2 Behavioural experiments

An important novelty of these experiments was the production of an acoustic environment that gave the experimental fish the opportunity to move away from the sound source and therefore to avoid high sound levels. This was possible due to the development of barriers that reduced sound transport and increased the sound pressure difference in the tank.

The unexpected preference for quarter 4 offered the possibility of testing the sound stimulus against a positive stimulus. This is comparable with the situation in a wind farm, where the fish will be confronted with a noisy area, but without disturbance due to fishing and providing the benefits of an artificial reef. Therefore the preference strengthens the outcome of the experiments. Additionally the larger fish numbers in tank quarter 4 made it possible to expose more fish to the sound than with equal fish distribution in the tank.

The results showed that the sound stimulus was not strong enough to cancel out the preference for quarter 4 in all fish but especially in cod it was strong enough to drive out a significant number of fish from the preferred area. The results also indicate that plaice detected the sound, but reactions were influenced by other stimuli.

5.1.2.1 Cod

Cod are hearing generalists with a hearing range from infrasound (ENGER et al. 1993) to about 470 Hz (CHAPMAN & HAWKINS 1973), with lowest hearing thresholds in a range from 60 to 380 Hz (SAND & HAWKINS 1973). Cod was chosen for the experiments as it is a commercially valuable gadoid species in the North and Baltic Sea. This species uses sound for intraspecific communication and therefore might also be affected by masking of biologically important signals. The hearing abilities of other species from the cod family such as haddock and lythe (*Pollachius pollachius*) were investigated by a number of authors (e.g. CHAPMAN & HAWKINS 1969, CHAPMAN & HAWKINS 1973, CHAPMAN 1973, OFFUT 1974 all summarized in NEDWELL et al. 2004) (Fig. 73). Given the similarity of the hearing abilities of these fish, it is suggested that the results obtained in these experiments can be transferred to other members of the cod family. The hearing ability of gadoids generally is better than in most other hearing generalists and therefore the reactions of other hearing generalists would not be expected to be more pronounced than reactions in cod.

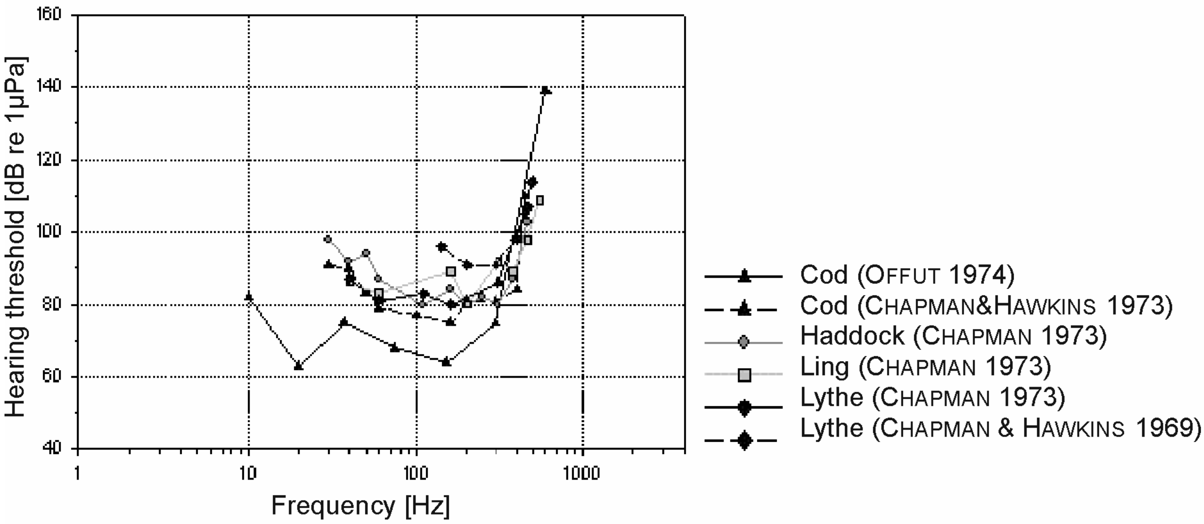


Fig. 73: Audiograms of gadoids from data of Nedwell et al. 2004

5.1.2.1.1 Behaviour of cod in the tank

The average number of active experimental cod was high at between 60% to 70%. A diel pattern with higher activity in daytime was more pronounced in juveniles. Resting was observed mostly in the vicinity of the water inflow. Higher fish numbers in quarter 4 during daylight periods were caused by the higher number of active fish. Cod swam frequently in smaller schools using the whole water column. The formation of schools influenced results because movements of a school caused sudden changes in fish numbers in the observed areas.

Feeding did not have a strong influence on the distribution of cod in the tank since most fish were active and all quarters were frequently visited. Although cod used the whole tank, a preference for tank quarter 4 was observed most of the time. An exception was the distribution of juvenile cod in the first week after introduction into the tank, when about 40% of the fish were present in quarter 1 and quarter 4 each on both sides of the long barrier.

5.1.2.1.2 Reaction of cod to sound

Looking at the numbers of fish in quarter 4 cod avoided the sound in many but not all experiments. The fine-scale movements of cod remaining inside the quarter indicated that those fish choosing to stay in the noisy quarter of the tank did avoid the direct vicinity of the sound source and therefore highest sound levels. SAND & HAWKINS (1973) determined the most sensitive hearing range of cod between 60 and 380 Hz. However in the experiments the detection of sound was not limited by the hearing thresholds but by the background noise level in the tank. With an average background noise level of about 105 dB re 1µPa and a signal-noise-ratio of 16 dB (CHAPMAN & HAWKINS 1973), cod should have been able to detect the frequencies at a sound level of about 121 dB re 1µPa in the most sensitive hearing range.

The results are mostly in accordance with the hearing ability of cod showing strongest reactions at the frequencies with lowest thresholds while the reactions were less pronounced at the lowest and highest tested frequencies. The reaction of cod to the frequency of 25 Hz was higher than expected, compared with the less pronounced reactions to 250 Hz despite a lower hearing threshold. But in cod the relevant stimulus at 25 Hz is the particle displacement (CHAPMAN & HAWKINS 1973). At the same sound pressure level the particle motion is higher in a tank compared with the open sea (CHAPMAN & SAND 1974) and therefore cod in the experiments were exposed to a higher stimulus at 25 Hz than they would have been in an offshore wind farm in the same sound pressure situation. The results showed stronger reactions of cod to frequencies up to 125 Hz with strongest reactions at the frequencies of 60 and 90 Hz. Since the stimulus at 25 Hz was not known but was expected to be higher than in the open sea the reactions to sound of 25 Hz were likely to be stronger in the experiments than in an offshore wind farm. Therefore a decrease of sound emission in the frequency range between 60 and 125 Hz seem to be most desirable.

During sound production in quarter 1 (contrary to all other experiments with sound production in quarter 4) the fish left quarter 1, which led to an increase in fish numbers in quarter 4. Apart from the two smallest fish in the experimental group, who returned and settled in quarter 1, fish mainly visited quarter 1 afterwards. On the other hand sound production in quarter 4 did not cause permanent avoidance of the area. This indicates that the acoustic stimulus was not strong enough to neutralize but to reduce the preference for quarter 4.

Considering these results, the sound emitted by offshore wind farms might be expected to cause at least temporary changes in distribution of cod. Habituation of cod to low frequency sound was observed by CHAPMAN (1976). Therefore habituation of cod to permanent sound seems conceivable if the wind farm area is attractive for the fish (see chapter 5.2).

Age groups

The experiments tested juveniles and adults separately since hearing ability is thought to change with maturity. ENGÅS et al. (1993) observed higher catch rates of juvenile cod during seismic shooting and suggested this might have been caused by adult fish moving away from the area as their hearing is more developed as a result of their larger swimbladders. On the other hand different authors (MCCARTNEY & STUBBS 1971, SAND & ENGER 1973, SAND & HAWKINS 1973) concluded that the bigger size of the swimbladder in adult cod would not improve the hearing ability in the frequency range to which cod is sensitive. The experiments in this study did not indicate a stronger reaction in the adult group. The absolute change in fish numbers in quarter 4 before and during sound production suggested a slightly stronger reaction in juvenile cod. However differences in reaction does not have to be related to the hearing ability but can be caused to the physiological state of the fishes and other factors like the season (MITSON 1995). Additionally juveniles that are more vulnerable to predators might be more wary of possible threats and therefore react stronger to the sound. In the experiments it was likely to be related to the less pronounced preference of juvenile cod for quarter 4 which might have caused a lower reaction threshold to the sound. Seasonal effects were ruled out by constant light and temperature conditions during all experiments.

Reaction thresholds

ENGÅS et al. (1993) calculated an avoidance threshold of cod to ship noise of between 110 and 120 dB re 1µPa. ENGÅS et al. (1996) observed reaction thresholds of 20 dB or less above detection threshold when cod and haddock were exposed to airgun noise. MITSON (2000) gave a threshold for avoidance reaction in herring and cod of 30 dB above hearing threshold. Another group of authors (NEDWELL et al. 1998 in NEDWELL et al. 2003a, NEDWELL et al. 2003b, NEDWELL & HOWELL 2004) expect slight behavioural reactions when the sound level is 70 dB above the hearing threshold of the species and avoidance behaviour would occur from a sound level 90 dB above hearing threshold. TURNPENNY & NEDWELL (1994) described reaction thresholds of different fish species to airguns at levels of 160 to 200 dB re 1µPa while other underwater sound sources to deter fish could already be effective at levels of 100 to 140 dB re 1µPa due to a better targeting of effective frequencies. The same study mentioned lower reaction thresholds in species of the cod family compared even with hearing specialist such as herring.

In the described experiments cod showed reactions to relatively low sound levels comparable with the results of ENGÅS et al. (1993) or even lower. The hearing ability of cod was influenced by the background noise in the tank that was about 105 dB re 1µPa on average in middle water which was higher than background noise in the North Sea for rather light wind conditions of up to 8 m s^{-1} (DEWI 2004).

CHAPMAN & HAWKINS (1973) determined a signal-noise-ratio of 16 dB and therefore cod would be able to detect the produced sound at a sound level of about 121 dB re 1µPa. From the results of ENGÅS et al. (1993) the avoidance threshold of cod would be 30-45 dB above detection threshold. This corresponds to 151 to 166 dB re 1µPa, which were reached in the vicinity of the sound source. The results point to even lower avoidance reaction thresholds in cod since the number of cod in quarter 4 decreased significantly in some experiments during sound production of 130 dB re 1µPa and therefore the sound levels hardly reached the thresholds given by ENGÅS et al. (1993) and MITSON (1995) even in the direct vicinity of the sound source.

The results of this study point to a far lower reaction threshold of cod to sound as the 90 dB above hearing threshold given from NEDWELL et al. (2003a) which was used to calculate for example an area of 5.5 km around pile driving activity (source level 260 dB re 1 µPa at 1 m) at the North Hoyle wind farm in which significant avoidance reactions of cod could occur. Using the calculation of NEDWELL et al. (2003a) on the results of the present experiments with lower reaction thresholds observed, the area in which avoidance reactions of cod would be expected would come to about 600 km around pile driving activity. ENGÅS et al. (1996) observed lower catch rates of cod and haddock in a distance of 18 nmi from an airgun (frequency band 60 to 310 Hz, peak level 249 dB re 1µPa) but pointed out that the area was too small to see the effects diminish with distance.

In experiments following on from the work presented in this thesis herring in a tank were exposed to playback sound of an 1.5-MW offshore wind turbine at a sound level of 135 dB re 1µPa (LÜDEMANN & MÜLLER 2006). The low hearing threshold of herring, 75 to 79 dB re 1µPa in a frequency range from 30 to 1200 Hz (ENGER 1967), was masked by the background noise level of 119 dB re 1µPa in the tank. Thus with a signal-noise-ratio of 15 to 20 dB, the produced sound was only just above the detection threshold of the experimental fish, but still produced significant avoidance reactions (LÜDEMANN & MÜLLER 2006).

By now the knowledge about hearing and reaction thresholds in fish to sound is too small to give reliable estimations and further research is urgently necessary.

5.1.2.2 Plaice

Plaice was chosen as a representative of the flat fish group which forms an important component of the fish community in North and Baltic Sea.

The auditory system of flat fish does not vary a lot and therefore the hearing ability of different species is expected to be comparable and the results of this experiment transferable to other flatfish species.

5.1.2.2.1 Behaviour of plaice in the tank

Juvenile plaice showed a strong preference for section D in quarter 4 where the water inflow was located whilst the preference was less obvious for adult plaice. This preference is most likely related to the natural habitats of these species. In the open sea juvenile plaice live in shallow waters exposed to strong currents caused by tides, weather and winds. With growing age the fish move to deeper and therefore stiller water (HEINCKE 1913).

The behaviour of plaice in the tank was in accordance with natural behaviour observed in the open sea (GIBSON 1980). The amount of activity, on average 25% at any one time in juveniles and 14% in adult plaice, was relatively high compared to field observations of 6% in wild juvenile plaice, but in accordance with other tank experiments under various conditions (GIBSON 1975).

Diel rhythm

Adult plaice in the experiments showed a strong diel rhythm closely related to the light regime, whilst juveniles exhibited a less pronounced pattern. The pattern only appeared over the course of some days after adjustment from the holding tank light regime, which mirrored seasonal daylight hours, to the experimental tank light regime which was held constant at 12 hours daylight.

The higher daytime activity can be related to the natural feeding activity observed during the day in the open sea (VERHEIJEN & DE GROOT 1967). However, a higher swimming activity in the water column at night, as described by VERHEIJEN & DE GROOT (1967) was not observed. GIBSON (1973) described two activity peaks in the very early morning (2-3 o'clock) and in the early evening. The first peak remained at the same time, whilst the second early evening peak shifted with the seasons (GIBSON 1973). VERHEIJEN & DE GROOT (1967) observed fish resting buried in the sand during the highest light levels in midday. The light level in the experiments remained constant during the light period and fish could not bury themselves. This may explain the relatively high activity levels throughout the day without distinct peaks. In addition to natural rhythms, higher activity in daytime periods might have been due to higher levels of external activity of humans around the tank during the day. Although the area directly surrounding the tank was blocked off, fish showed a reaction to people who stopped close to the tank, probably expecting food. Although this kind of disturbances were mostly avoided it cannot be ruled out that higher activity in the tank room in daytime influenced the activity level of plaice in the tank.

In the wild, the rhythm of plaice in coastal areas is also connected to the tides with higher activity before low tide when the fish have to move with the ebbing water to avoid stranding

(GIBSON 1976). In the experiments the fish were kept in the holding tank for a longer period of time before the beginning of the experiment. Therefore a tidal rhythm was not expected and could not be observed. GIBSON (1976) also observed that the tidal rhythm was replaced by a diel rhythm after only a few days in an indoor tank. In the natural environment, as plaice mature they shift from near shore waters to deeper waters without tidal *zeitgeber* and this accounts for the quick adaptation (GIBSON 1976).

5.1.2.2.2 Reaction of plaice to sound

Plaice are hearing generalists with high hearing thresholds in a relatively narrow frequency band from infrasound to about 250 Hz (CHAPMAN & SAND 1974, KARLSEN 1992a). Therefore it was not expected that they would show strong reactions to sound as emitted by offshore wind farms.

From a number of the experiments, including the habituation with sound investigation, it appeared that plaice were able to detect the sound produced but showed ambiguous reactions that were probably related to a combination of stimuli in addition to sound.

The relevant stimulus for hearing in flatfish is the particle motion (HAWKINS & MACLENNAN 1975, KARLSEN 1992a). Levels of particle motion could not be determined in the tank, but it was expected that particle movement was higher in quarter 4 containing the sound source. CHAPMAN & SAND (1974) pointed out that the particle motion in a tank is higher than it is in open water and that this could lead to a wider frequency range in the hearing of fish. The authors also suggested that for this reason the reaction of fish to sound in a tank could be higher than it would be under natural conditions. While the situation in a tank makes higher particle motion likely, the tank bottom could be considered as a rather rigid boundary that would lead to lower particle acceleration in its vicinity (FREYTAG 1967), compared with the softer sediment of the sea floor where particle movement increases close to the boundary. This effect would be important for plaice, which spend most of their time resting at the bottom.

From the audiogram of plaice as given by CHAPMAN & SAND (1974) and KARLSEN (1992a) (Fig. 7) stronger reactions would have been expected to sound of 60 to 125 Hz, since the hearing threshold at these frequencies is lower compared with the frequencies of 25 and 250 Hz. Both increases or decreases in fish numbers were observed during sound production, without a clear pattern related to frequency, sound level or experimental group.

LAGARDERE et al. (1994) tested another flatfish species, sole (*Solea solea*) in a large, shallow (1m) seawater pond and found changes in distribution which were related to low frequency sound originating from wind. Significant differences were found depending on the strength and direction of the wind with sole preferring the less noisy areas. The authors suggested that sole used an acoustic gradient for orientation and that a minimum difference of 6 dB was necessary between areas to cause behavioural changes. The wind induced sound was at frequencies of between 40 to 140 Hz, with highest noise levels of up to about 100 dB re 1µPa between 60 and 90 Hz (LAGARDÈRE et al. 1994). In the results presented in this study, at 60 Hz, attraction rather than avoidance behaviour was observed.

Sensitivity to sound depends not only on the produced sound but also on the developmental stage and physiological state of the fish (POPPER 2003). However, no clear difference in behaviour between juvenile and adult plaice was observed in these experiments.

The sound levels tested were comparable with sound levels emitted by existing turbines and with predictions for larger turbines planned for future wind farms (DEWI 2004). The lack of a consistent reaction to the different frequencies means that recommendations concerning reduction of particular frequencies is not possible. However, a permanent avoidance of offshore wind farms would not to be expected if the area itself is attractive to plaice.

5.1.2.2.3 Habituation of plaice to sound

A delay of settlement and alteration of the distribution in quarter 4 was observed when the fish were inserted into the tank during sound production compared with settlement without sound. The results show that sound did not prevent settlement of juvenile plaice in the noisy environment of quarter 4.

A period of about 30 hours was required to become acclimatized to the noise. It would therefore be expected that plaice would habituate to sound produced by an offshore wind farm if the area itself is attractive enough to fish. However, results should be treated with care, since they are based on only a small number of fish in the experiments. While the two groups of fish during adaptation without sound contained 20 fishes each, only 6 fish were used in the habituation with sound experiment. Therefore individual preferences may have had a greater influence on the results.

5.2 Potential effects of offshore wind farms on fish

The results show that cod and plaice were capable of detecting the sound and that cod showed significant avoidance behaviour to most of the produced sound fields with strongest reactions to sound between 60 Hz and 125 Hz. In plaice a clear pattern related to frequency could not be observed. Although the sound stimulus was strong enough to cause significant lower numbers of cod in the quarter of sound production it did not scare away all fish from the generally preferred area. The stronger reaction of cod is in accordance with the better hearing ability compared with the poor hearing ability of plaice. Clear differences between age groups were not observed.

From the results it would be expected that the sound emitted by offshore wind farms could cause at least temporary changes in distribution of cod and plaice. In plaice habituation to sound could be observed in an additional experiment but results about possible habituation of cod could not be obtained. From the strengths of the reaction to the sound a permanent avoidance of the wind farm area would not be expected since even in the experiments with strongest reactions only a part of the fish group left the noisy area.

The preference for quarter 4 in the tank might be compared to positive effects caused by wind farms such as artificial reef effects (see chapter 1.2.4.4) and the fact that fishery will not take place in wind farms. For these reasons wind farms would effectively provide a refuge for fish. Therefore the results indicate that the sound produced by offshore wind farms will not be strong enough to superimpose the positive effects mentioned. Consequently the potentially negative effects of sound emission and electro-magnetic fields may be reduced by positive effects of the wind farm. The balance of these effects is currently unknown.

Further research on masking of biologically important sounds and reaction thresholds is urgently necessary to estimate the area around a wind farm in which the behaviour of fish will be influenced and in order to fully understand possible impacts.

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Appendix

List of Figures and Tables

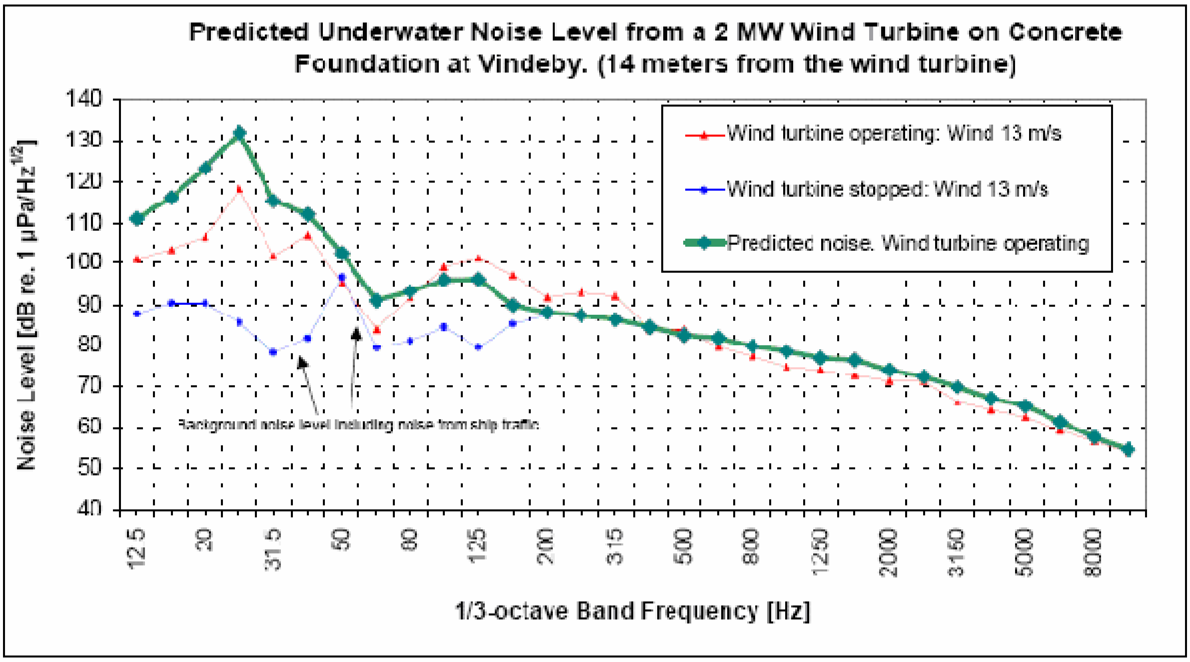

Fig. A 1: Measured and predicted sound emissions of offshore wind turbines at a concrete foundation. Fig. from Degn (2000).

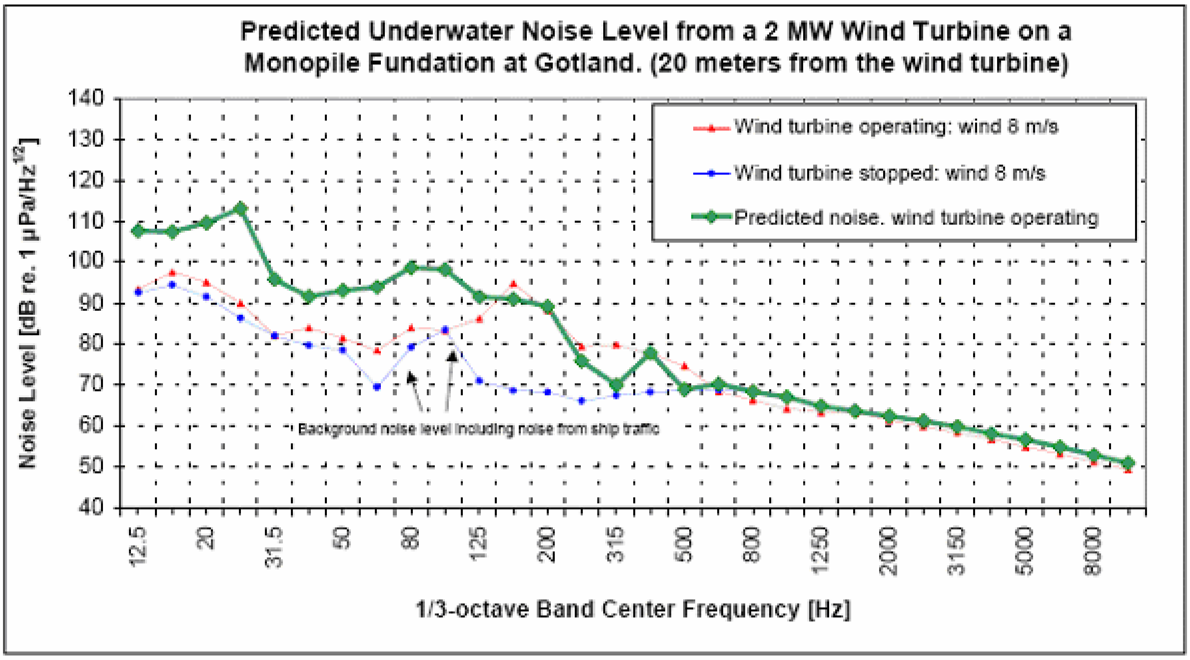

Fig. A 2: Measured and predicted sound emissions of offshore wind turbines at a monopile foundation. Fig. from DEGN (2000).

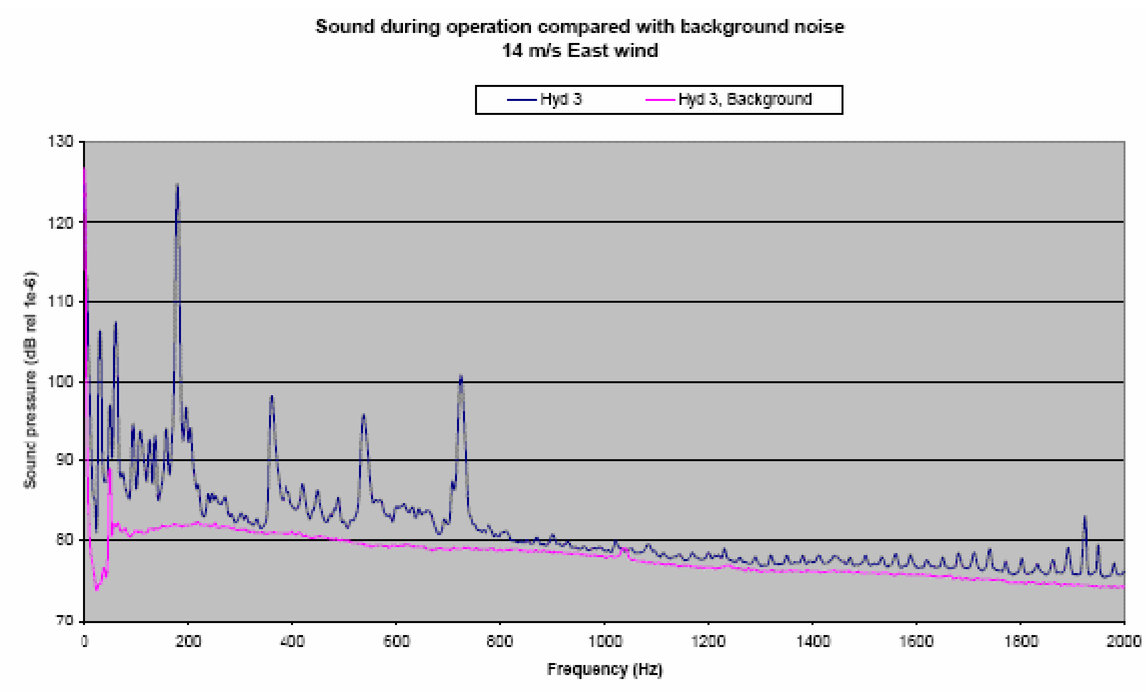

Fig. A 3: Sound measurements at a distance of 83 m from a 1,5-MW offshore wind turbine. Figure from INGEMANSSON (2003).

Table A 1: Offshore wind farms mentioned in the thesis

Wind farm	Country	status	number and size of turbines	in operation since
Horns Rev	Denmark	i.o.	80 x 2MW	2002
North Hoyle	UK	i.o.	30 x 2 MW	2004
Nysted (R ødsand)	Denmark	i.o.	70 x 2.2 MW	2003
Robin Rigg	Scotland	pl.	60 x 3 MW	announced for 2009
Svante	Sweden	i.o.	1 x 220 kW	1990
Utgrunden	Sweden	i.o.	7 x 1.5 MW	2001
Vindeby	Denmark	i.o.	11 x 450 kW	1991

pl. = planned,   i.o. = in operation

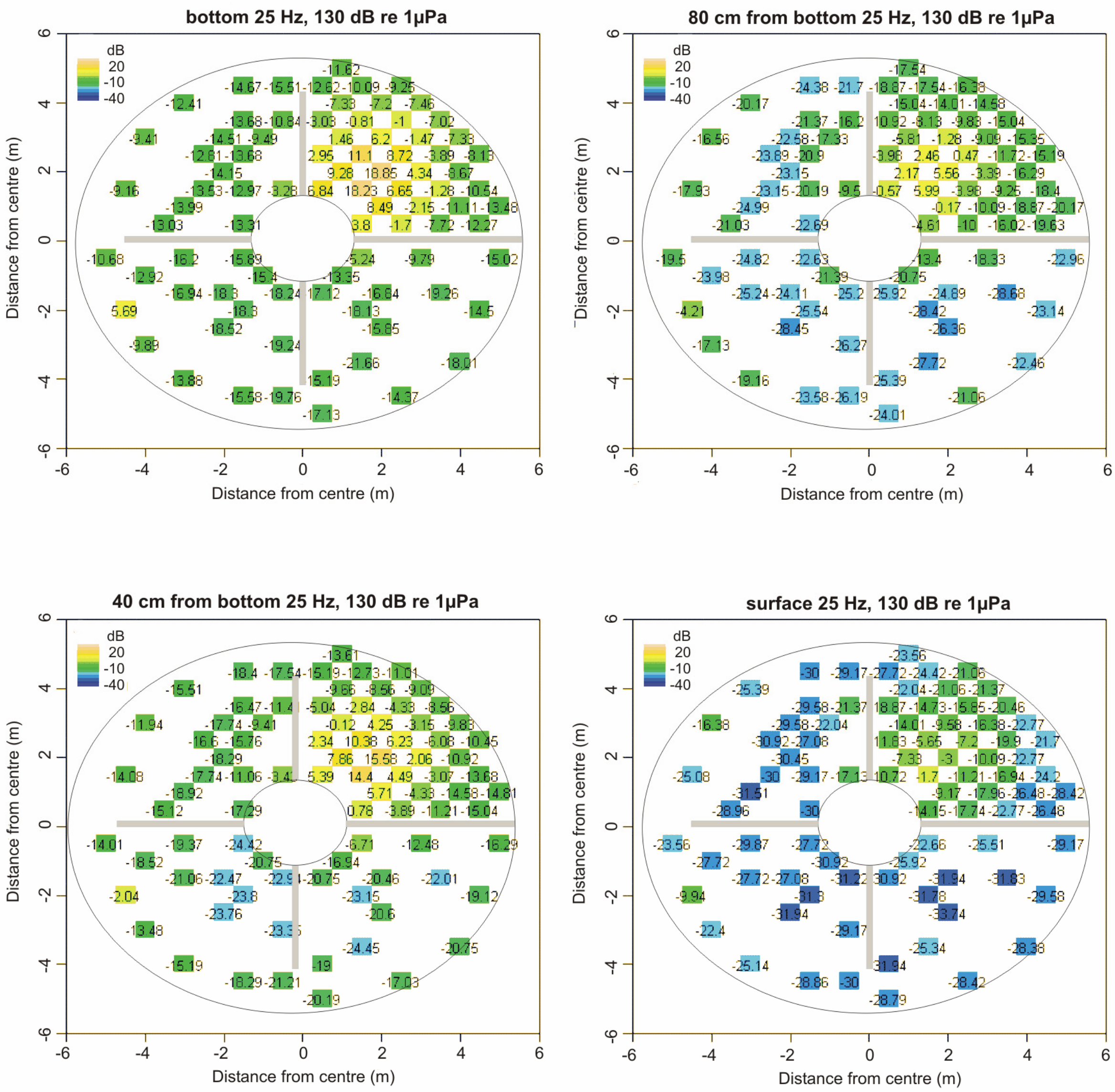

Fig. A 4: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 25 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

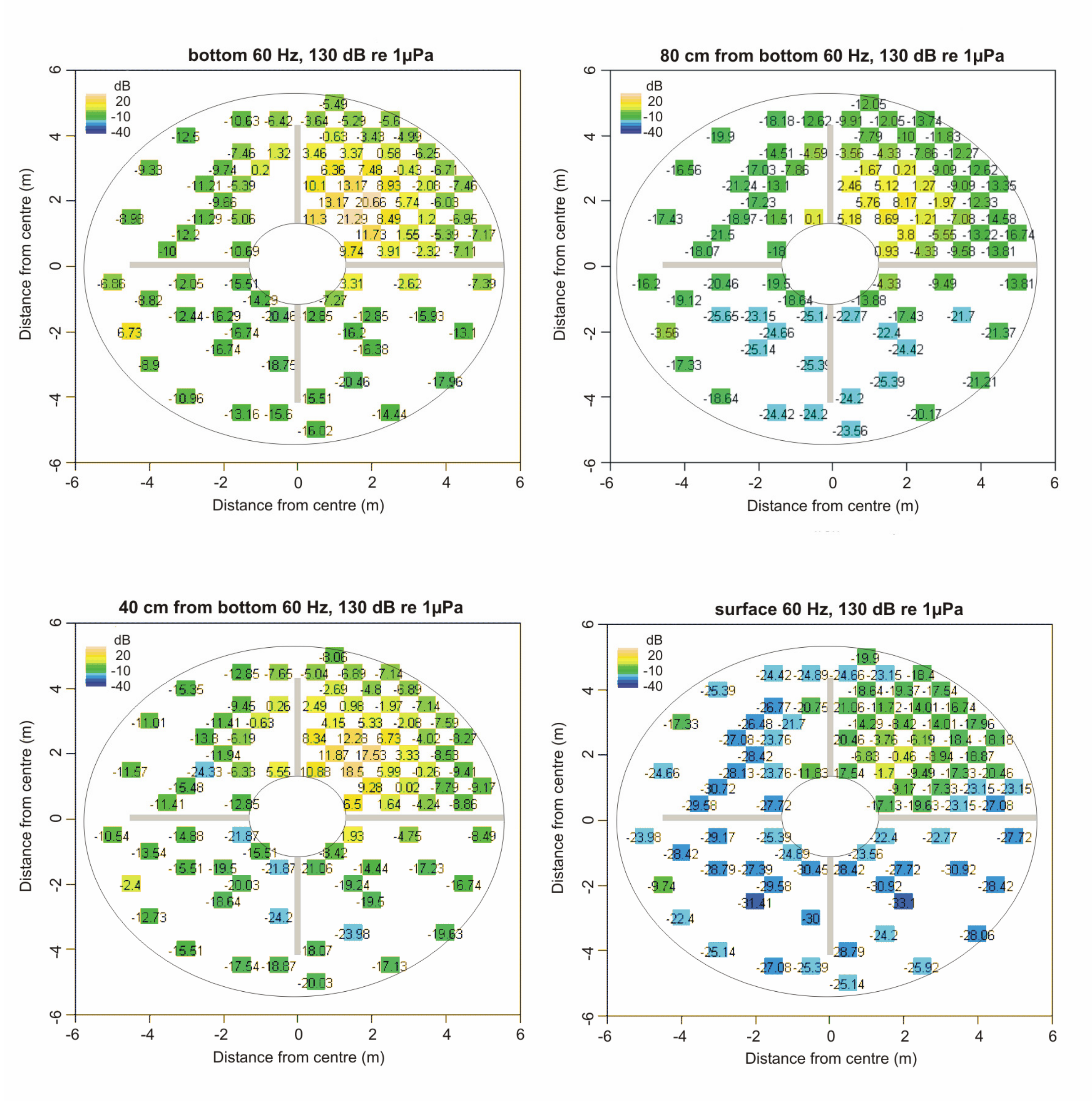

Fig. A 5: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 60 Hz and a sound level of 130 dB re 1μPa. The measurements are presented as the difference from the produced sound level.

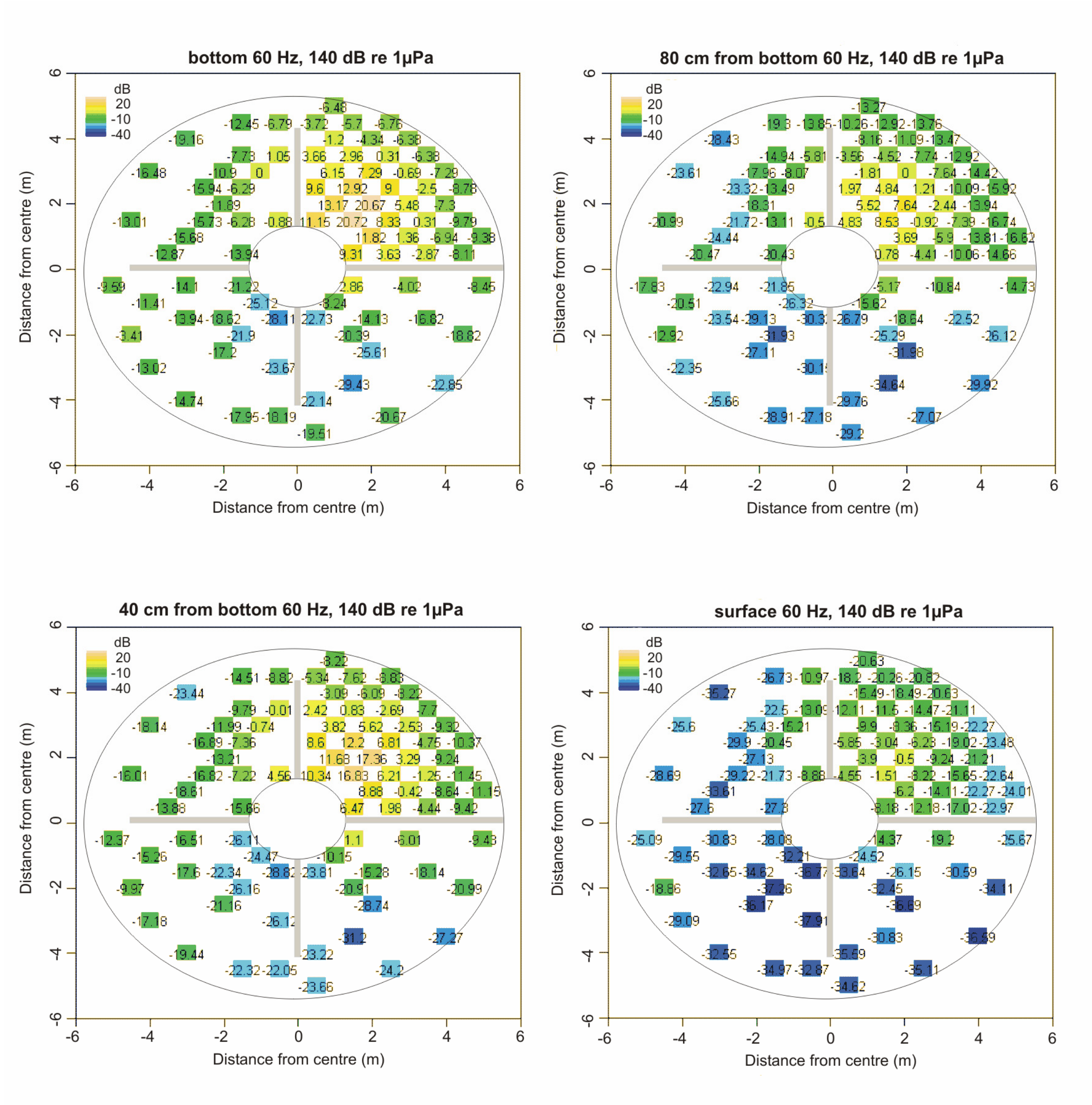

Fig. A 6: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 60 Hz and a sound level of 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

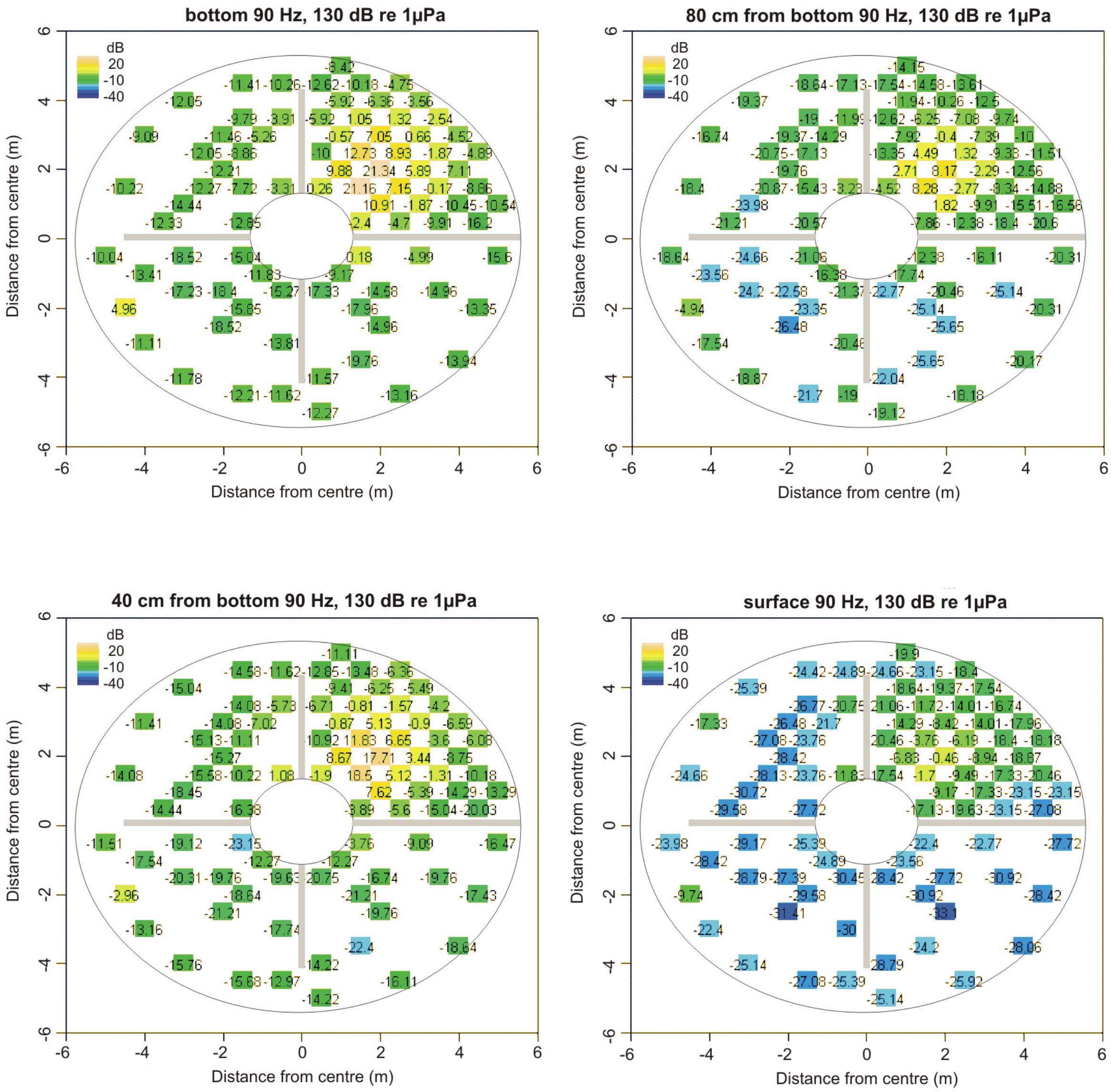

Fig. A 7: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 90 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

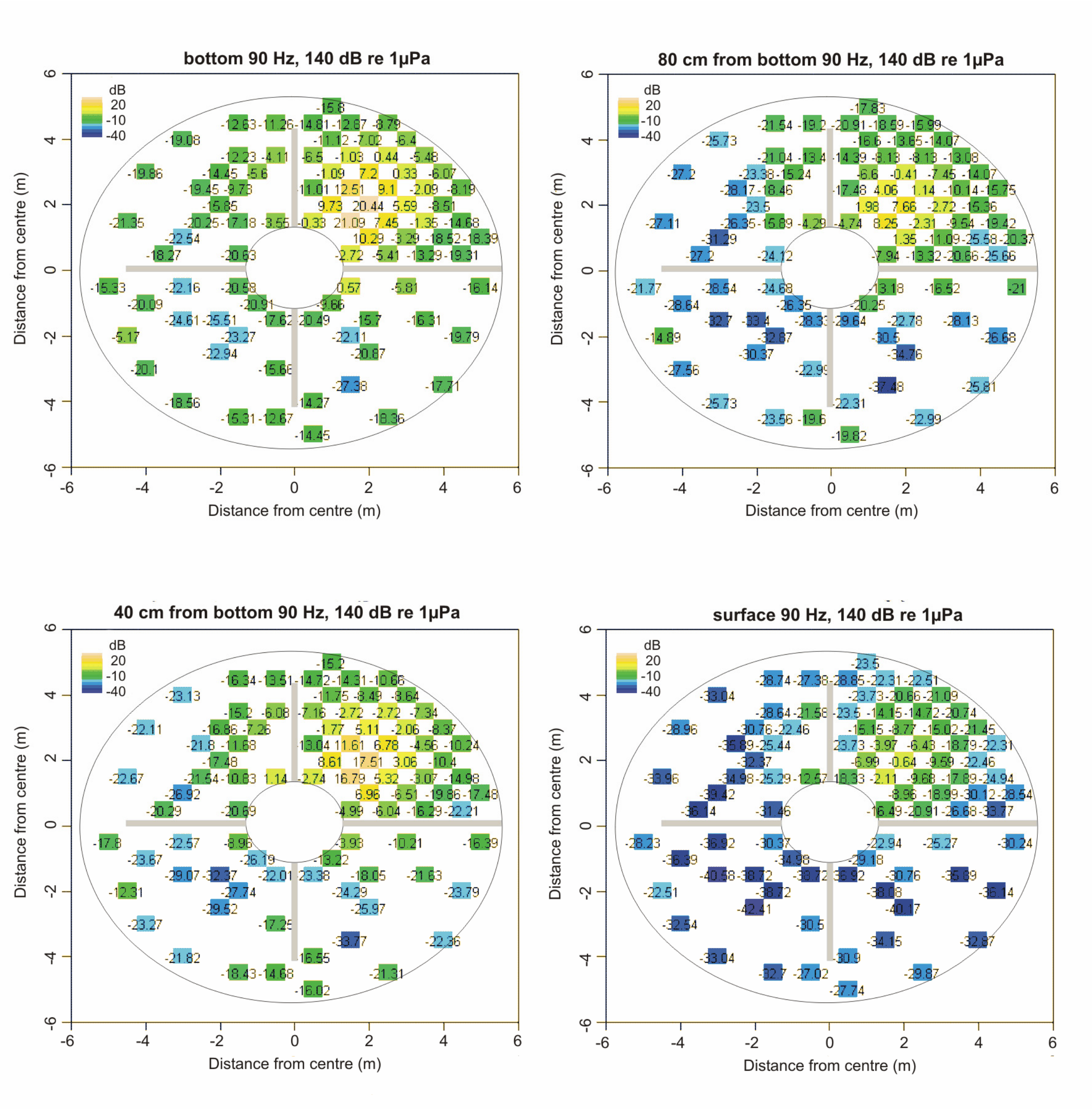

Fig. A 8: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 90 Hz and a sound level of 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

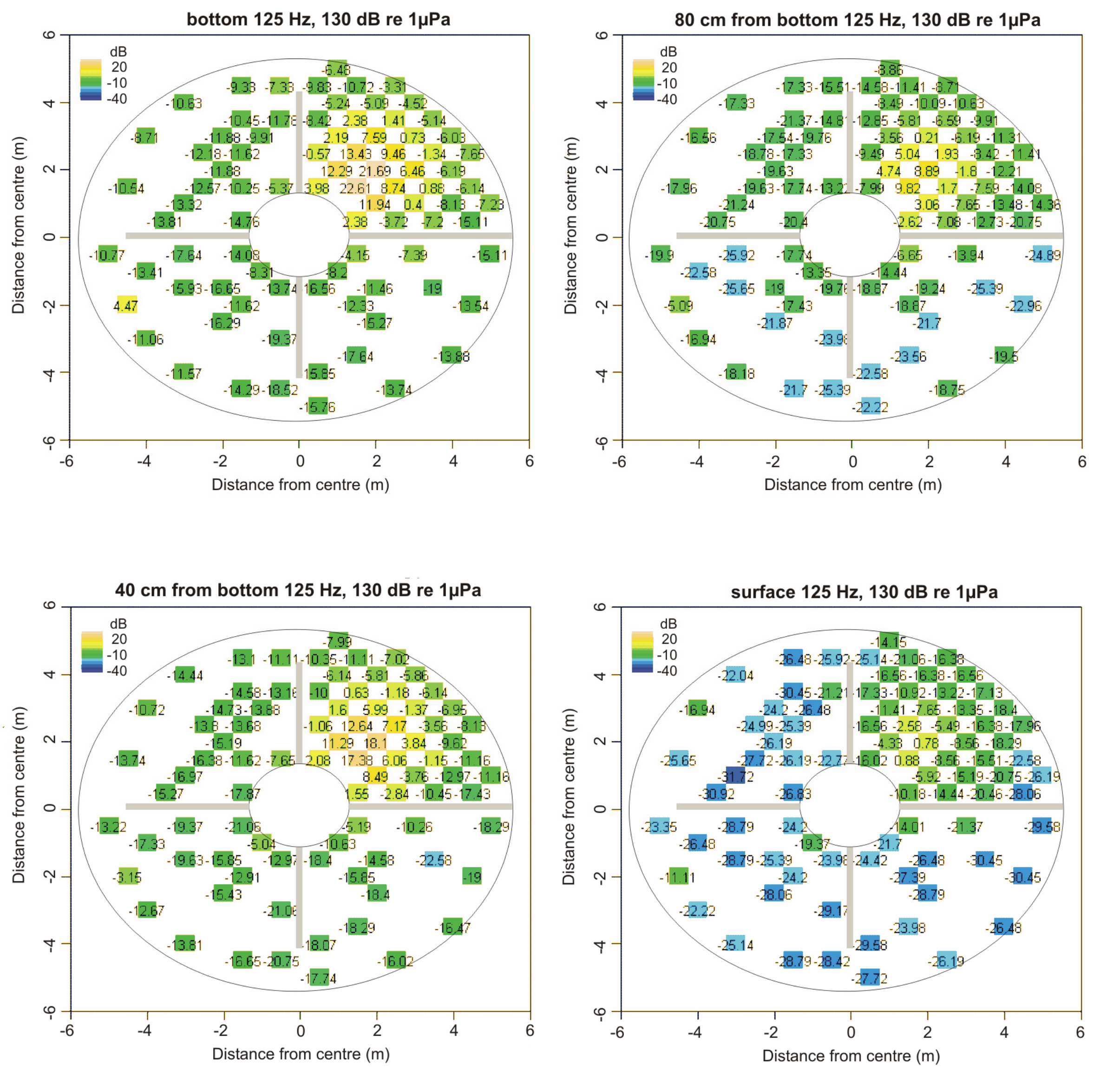

Fig. A 9: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 125 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

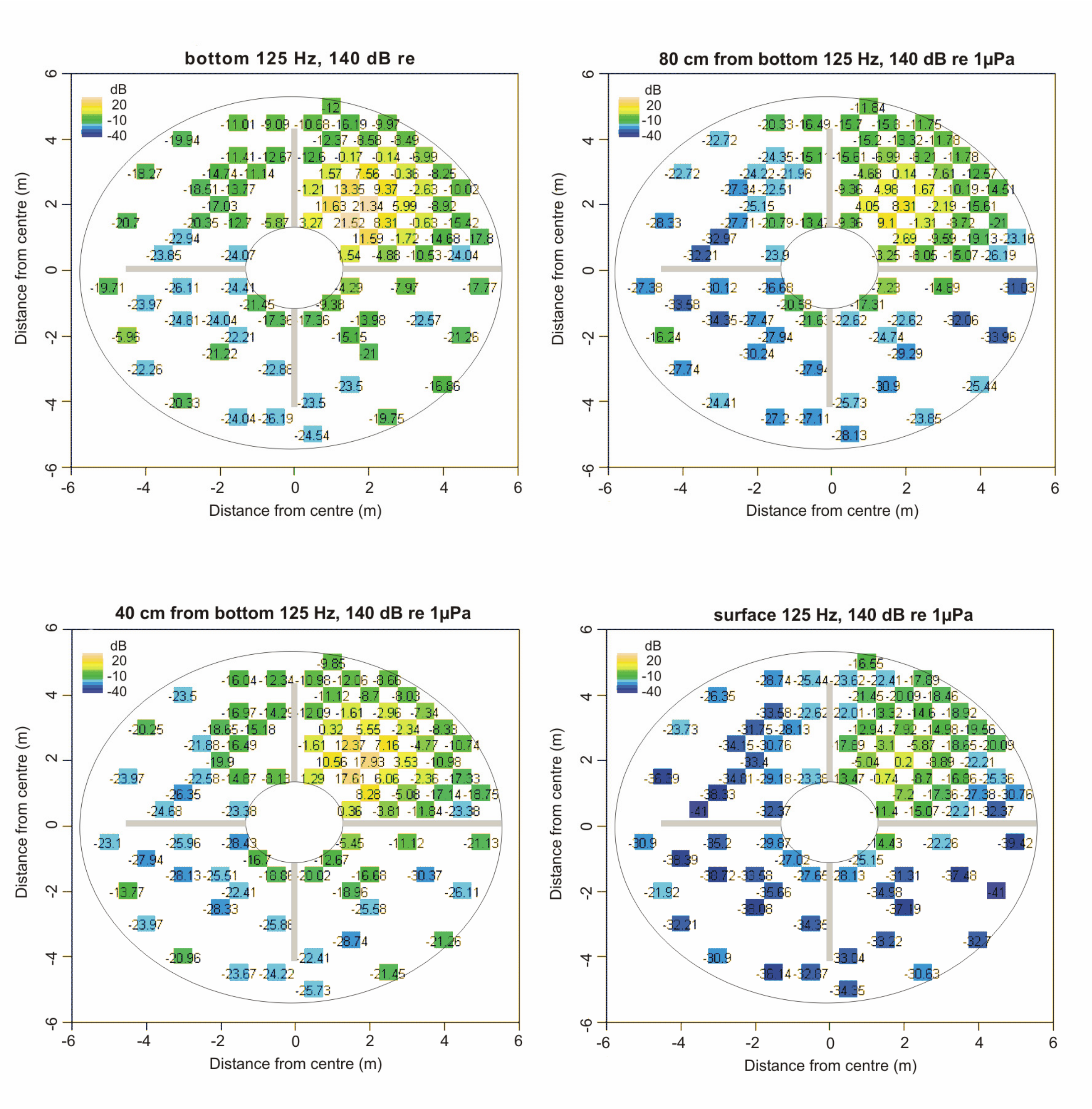

Fig. A 10: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 125 Hz and a sound level of 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

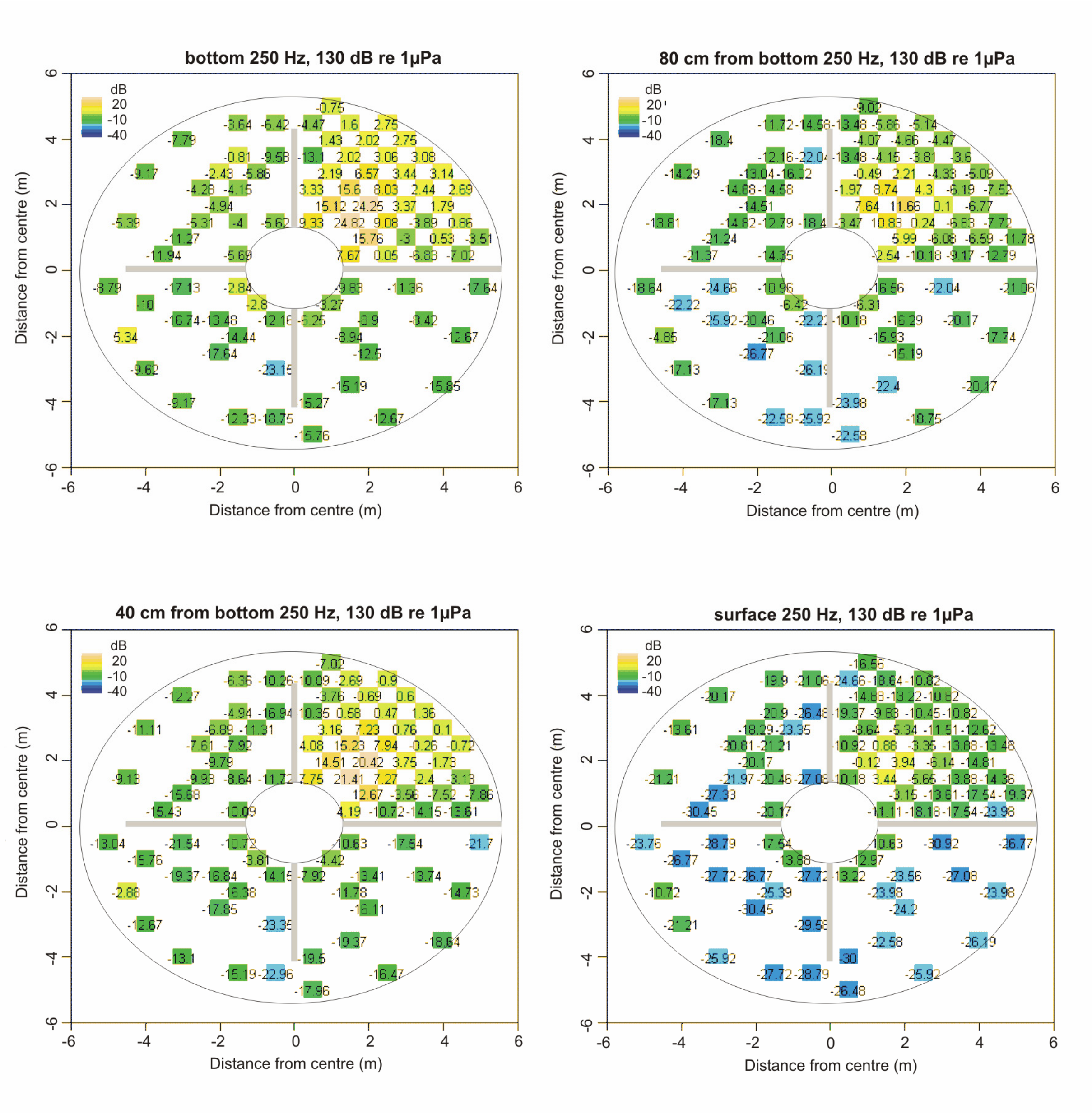

Fig. A 11: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 250 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

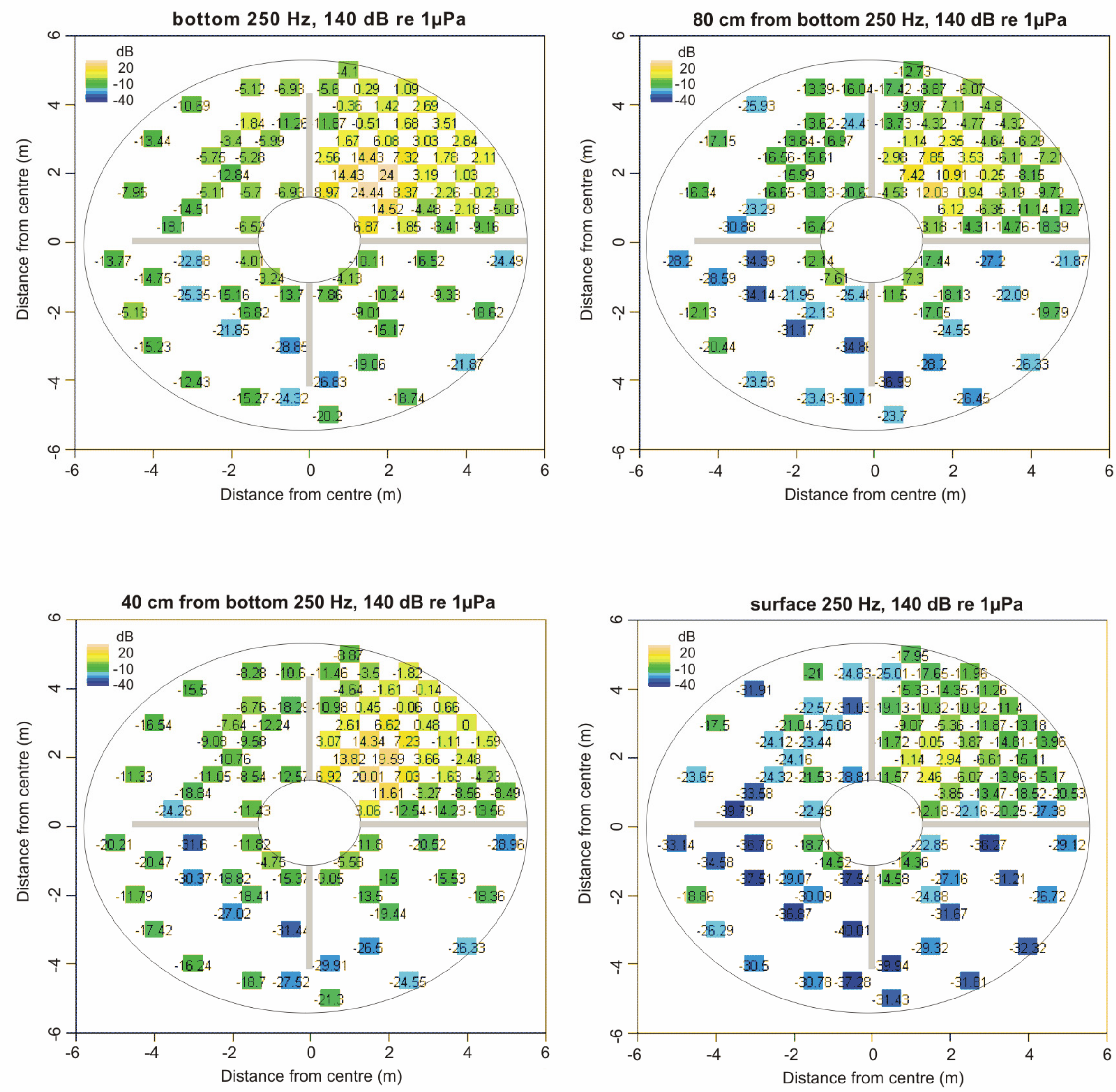

Fig. A 12: Sound field in the tank divided by barriers during sound production in tank quarter 1 at a frequency of 250 Hz and a sound level of 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

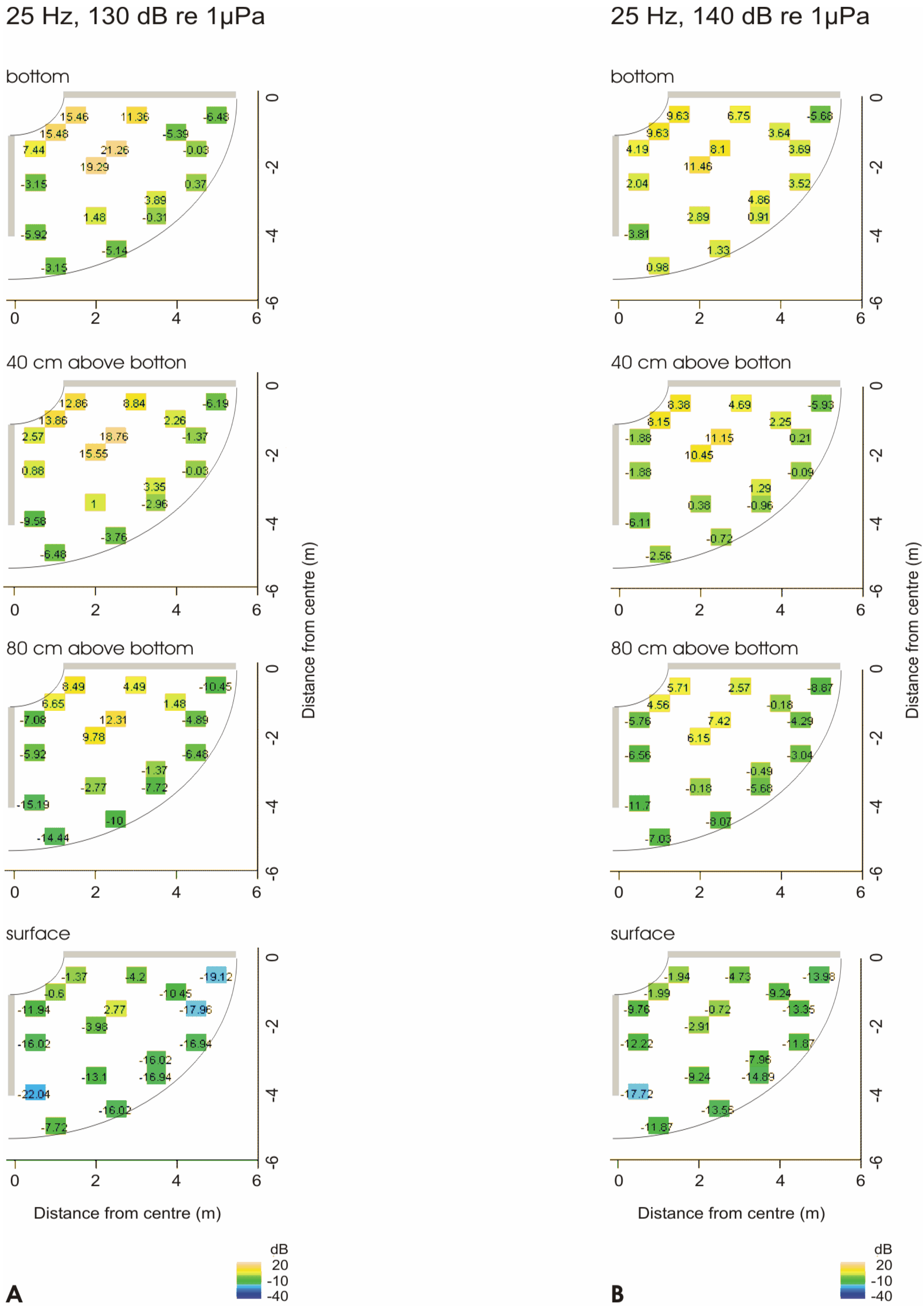

Fig. A 13: Sound field in tank quarter 4 during sound production in quarter 4 at a frequency of 25 Hz and a sound level of A: 130 dB re 1µPa and B: 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

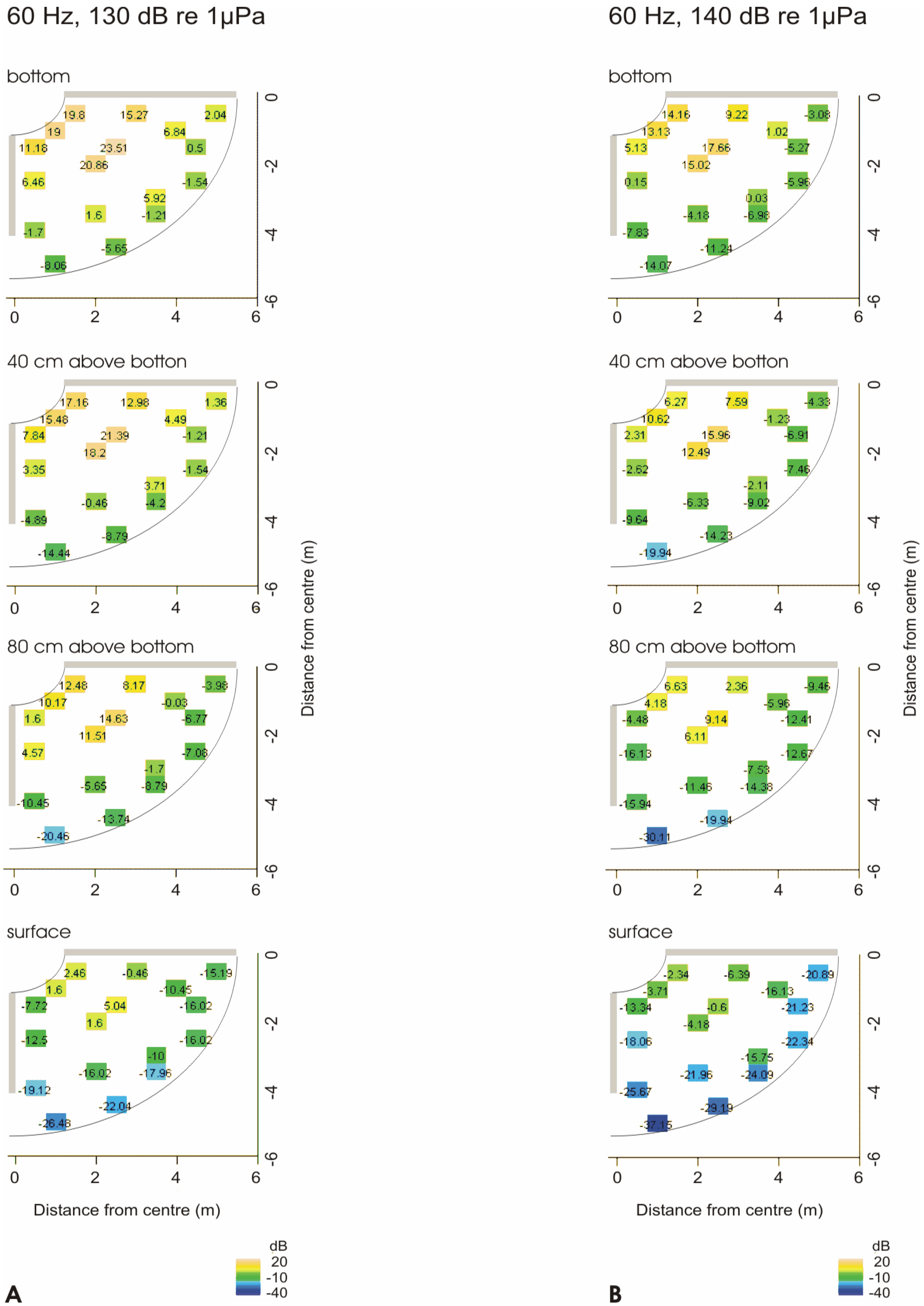

Fig. A 14: Sound field in tank quarter 4 during sound production in quarter 4 at a frequency of 60 Hz and a sound level of A: 130 dB re 1µPa and B: 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

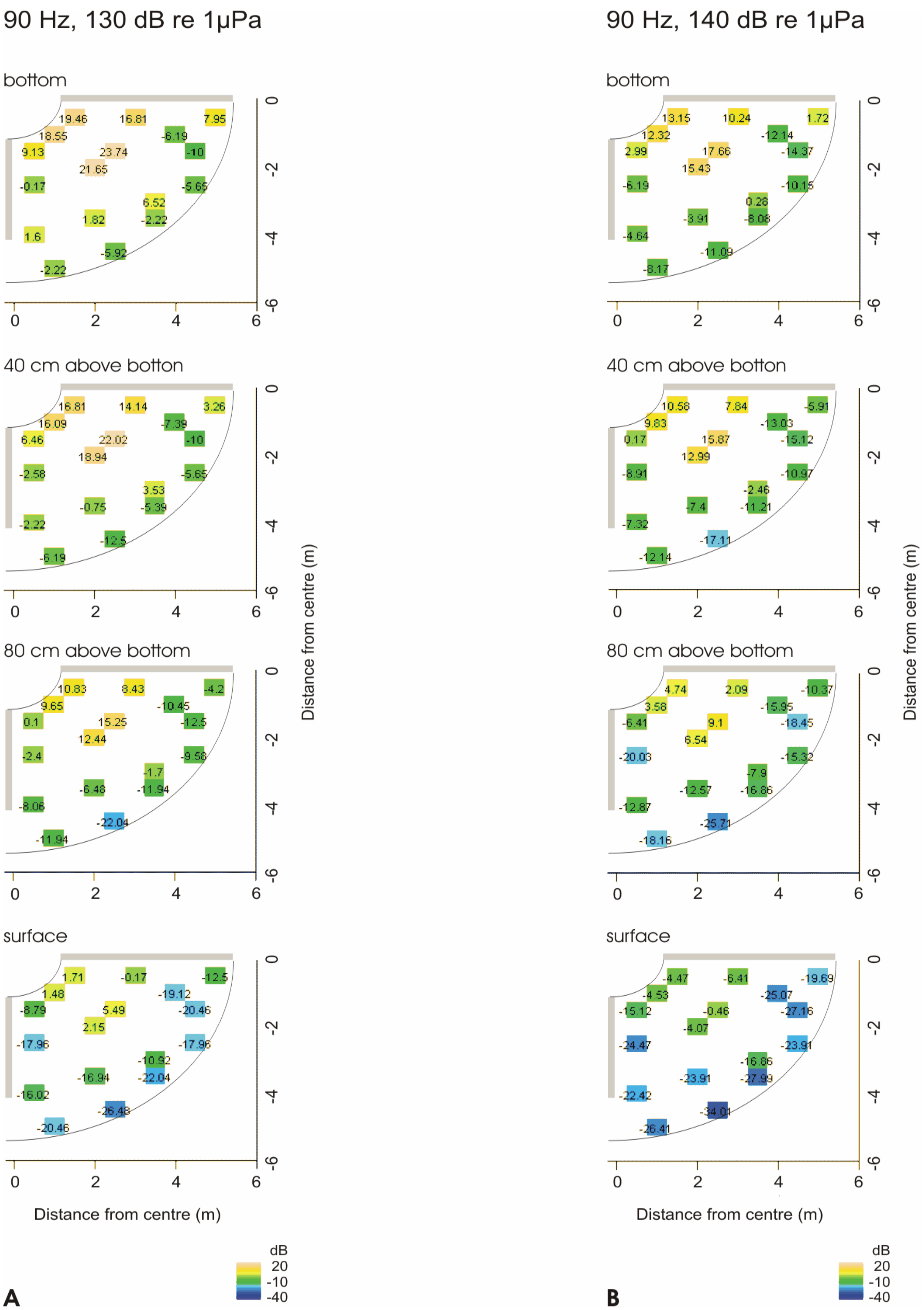

Fig. A 15: Sound field in tank quarter 4 during sound production in quarter 4 at a frequency of 90 Hz and a sound level of A: 130 dB re 1µPa and B: 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

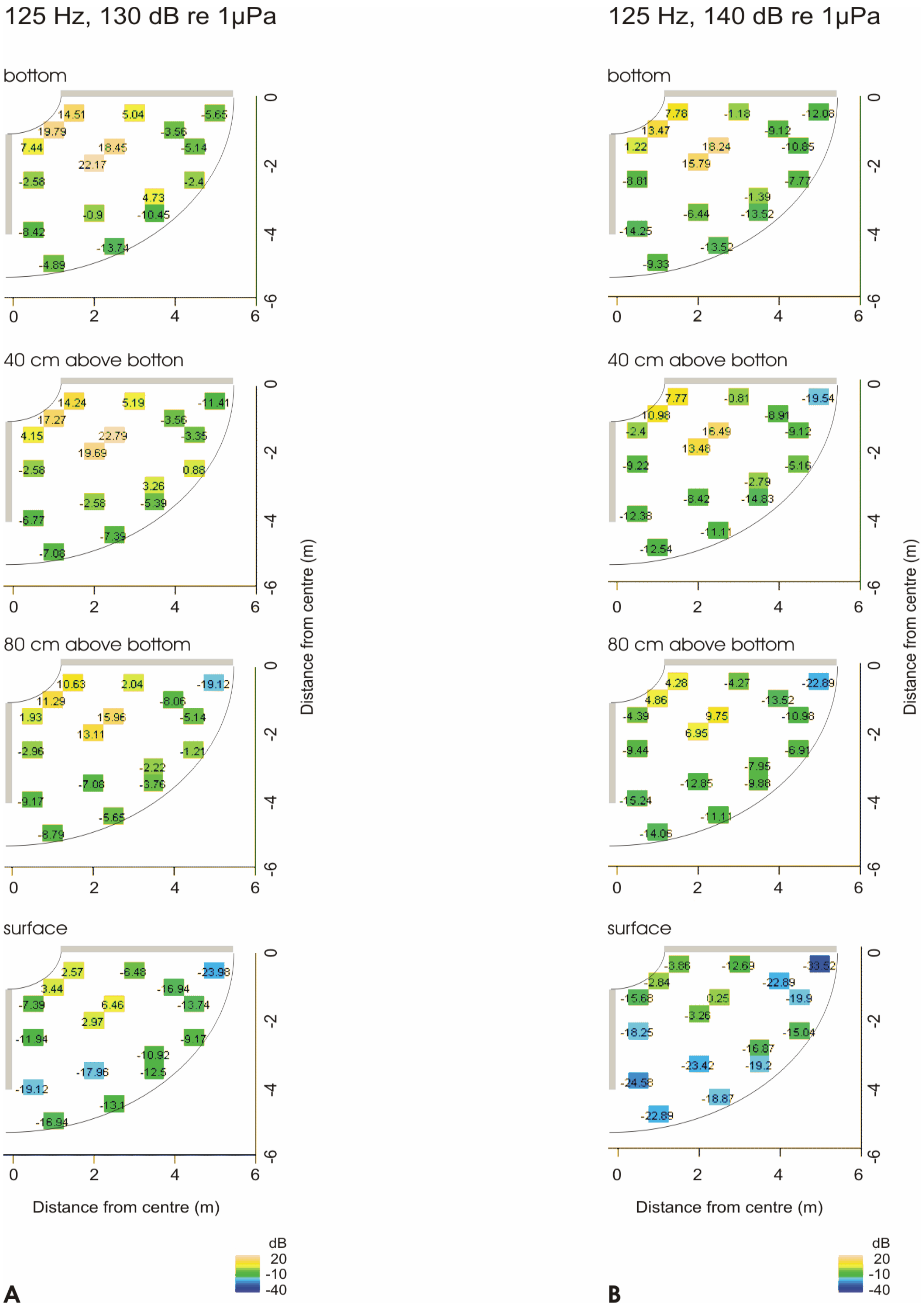

Fig. A 16: Sound field in tank quarter 4 during sound production in quarter 4 at a frequency of 125 Hz and a sound level of A: 130 dB re 1µPa and B: 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

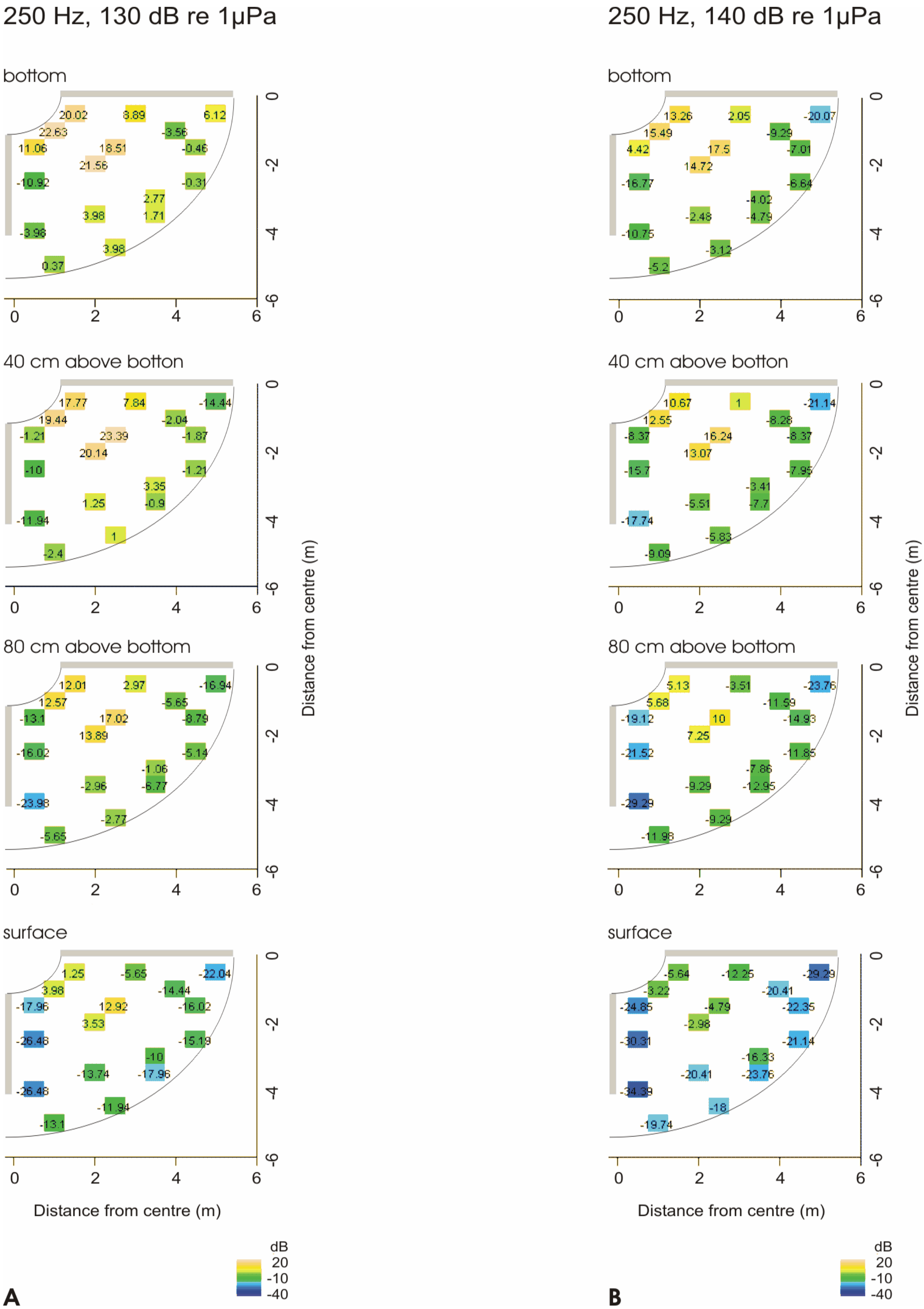

Fig. A 17: Sound field in tank quarter 4 during sound production in quarter 4 at a frequency of 250 Hz and a sound level of A: 130 dB re 1µPa and B: 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

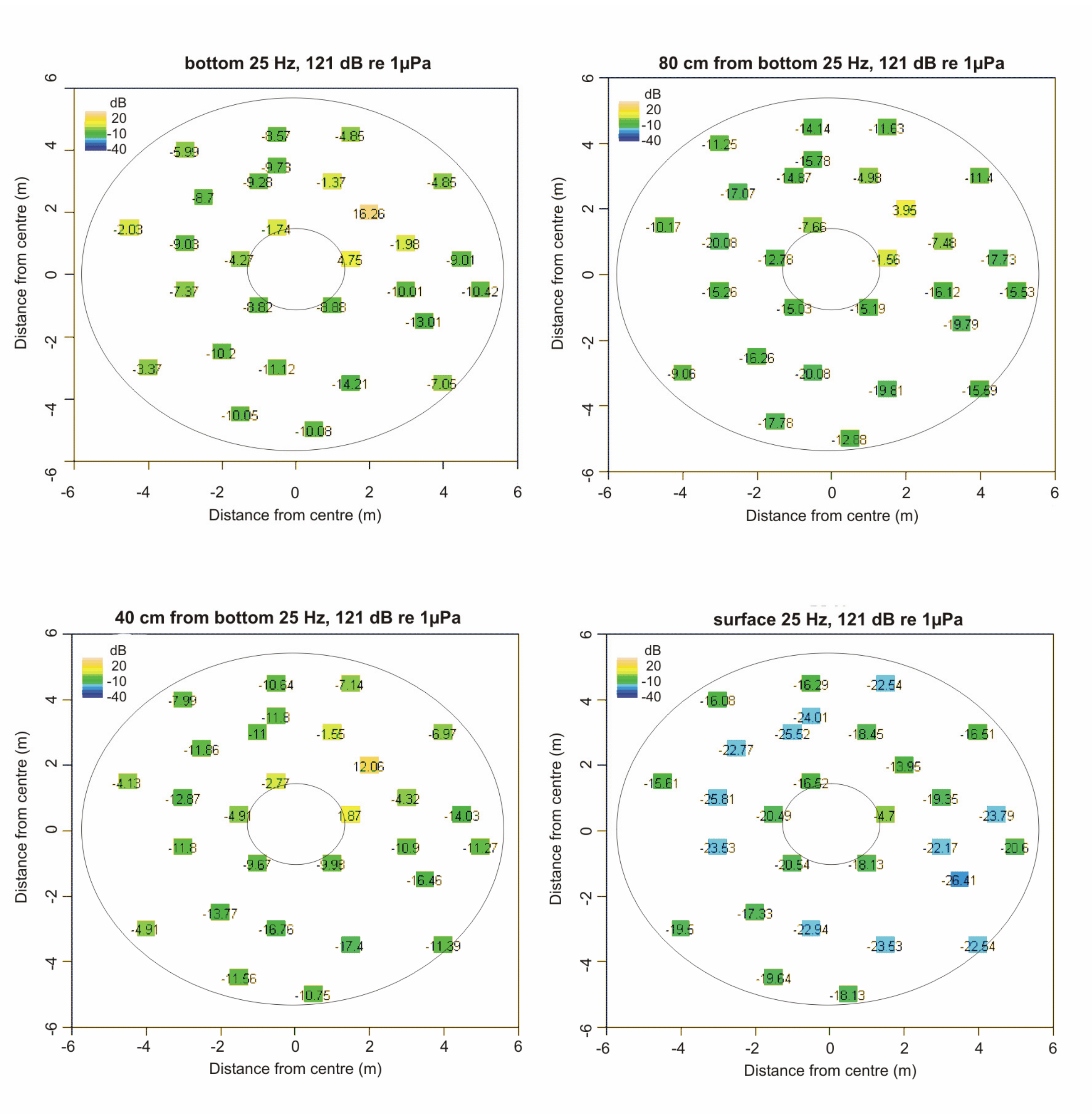

Fig. A 18: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 25 Hz and a sound level of 121 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

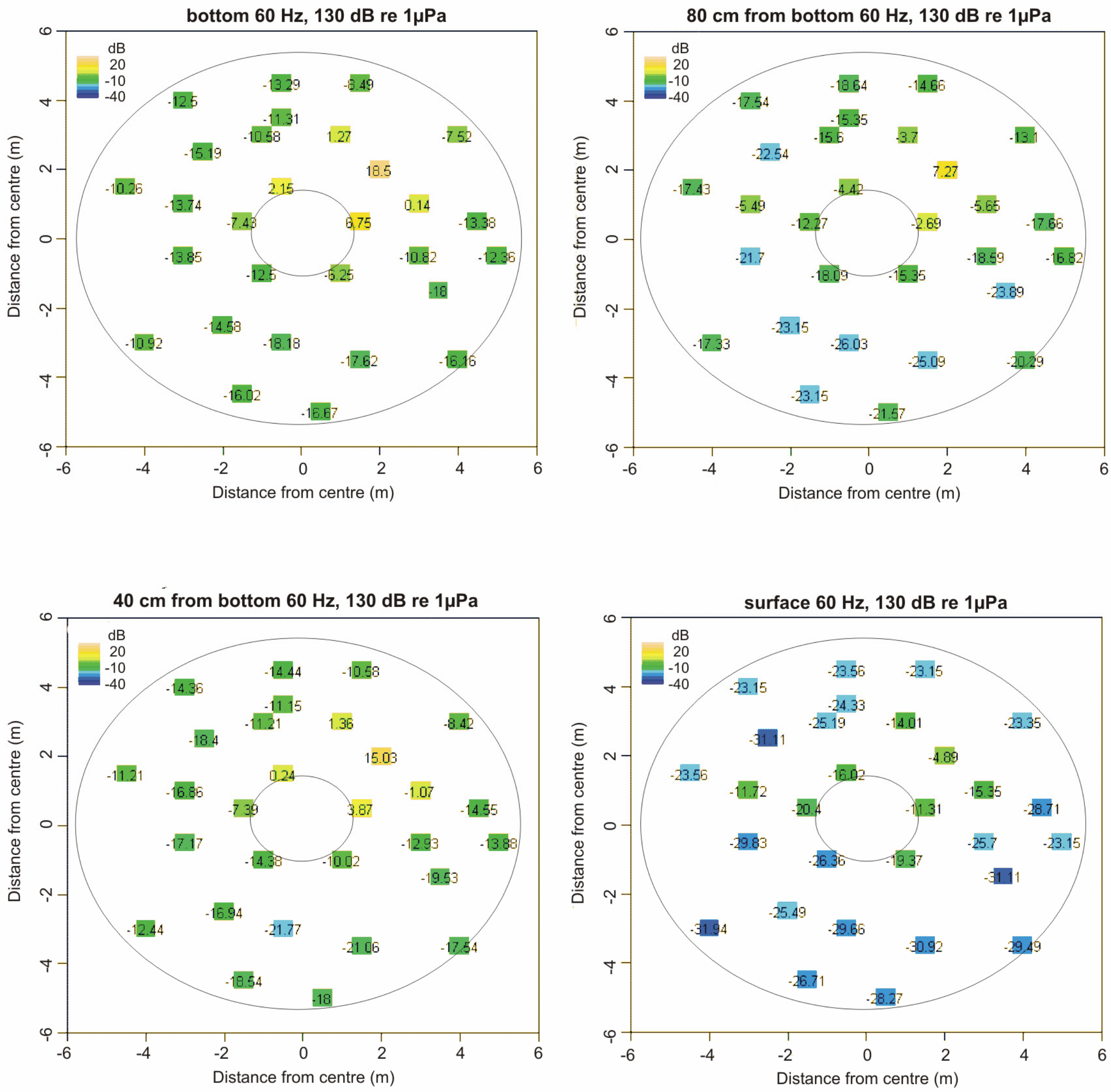

Fig. A 19: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 60 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

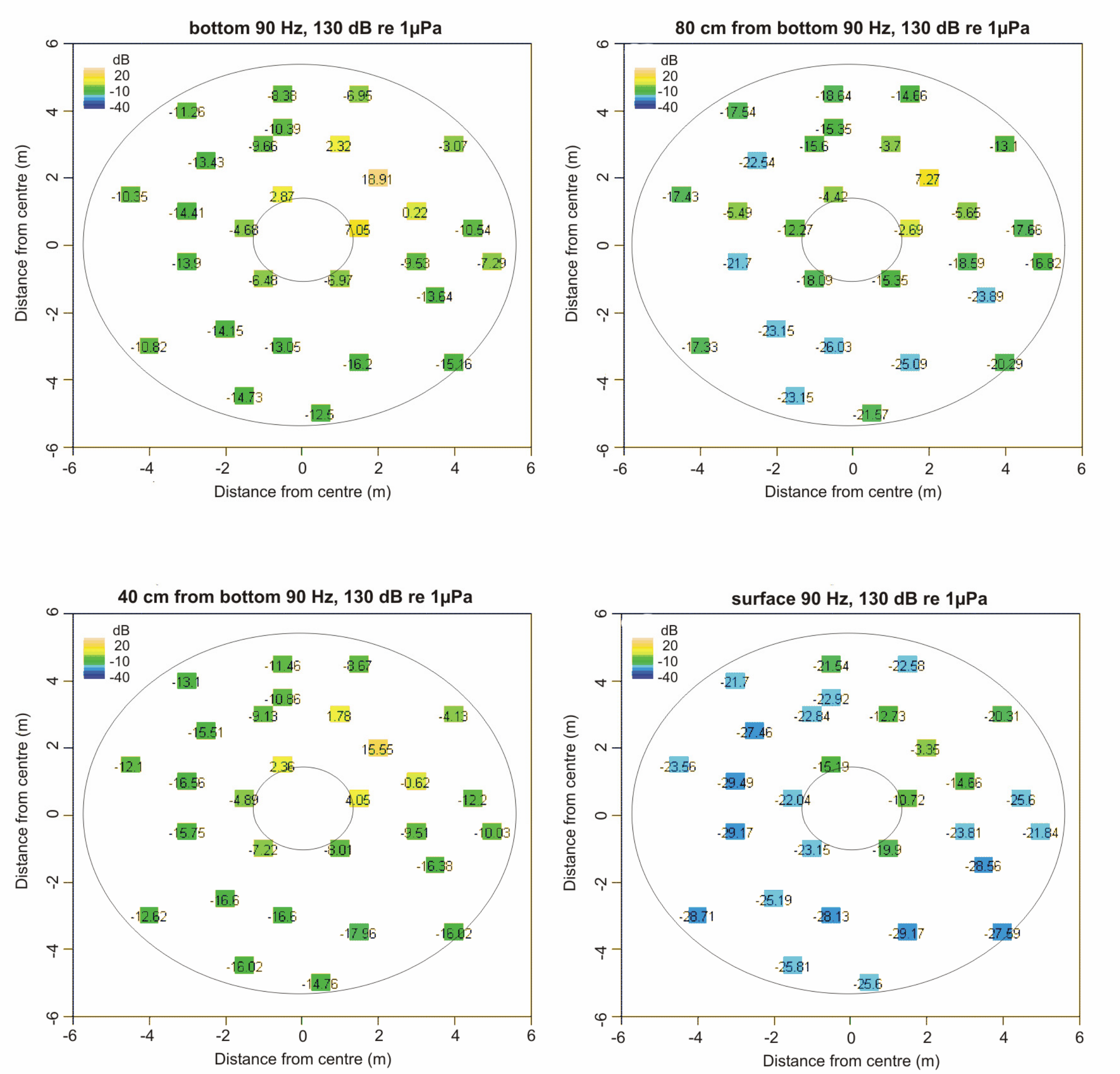

Fig. A 20: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 90 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

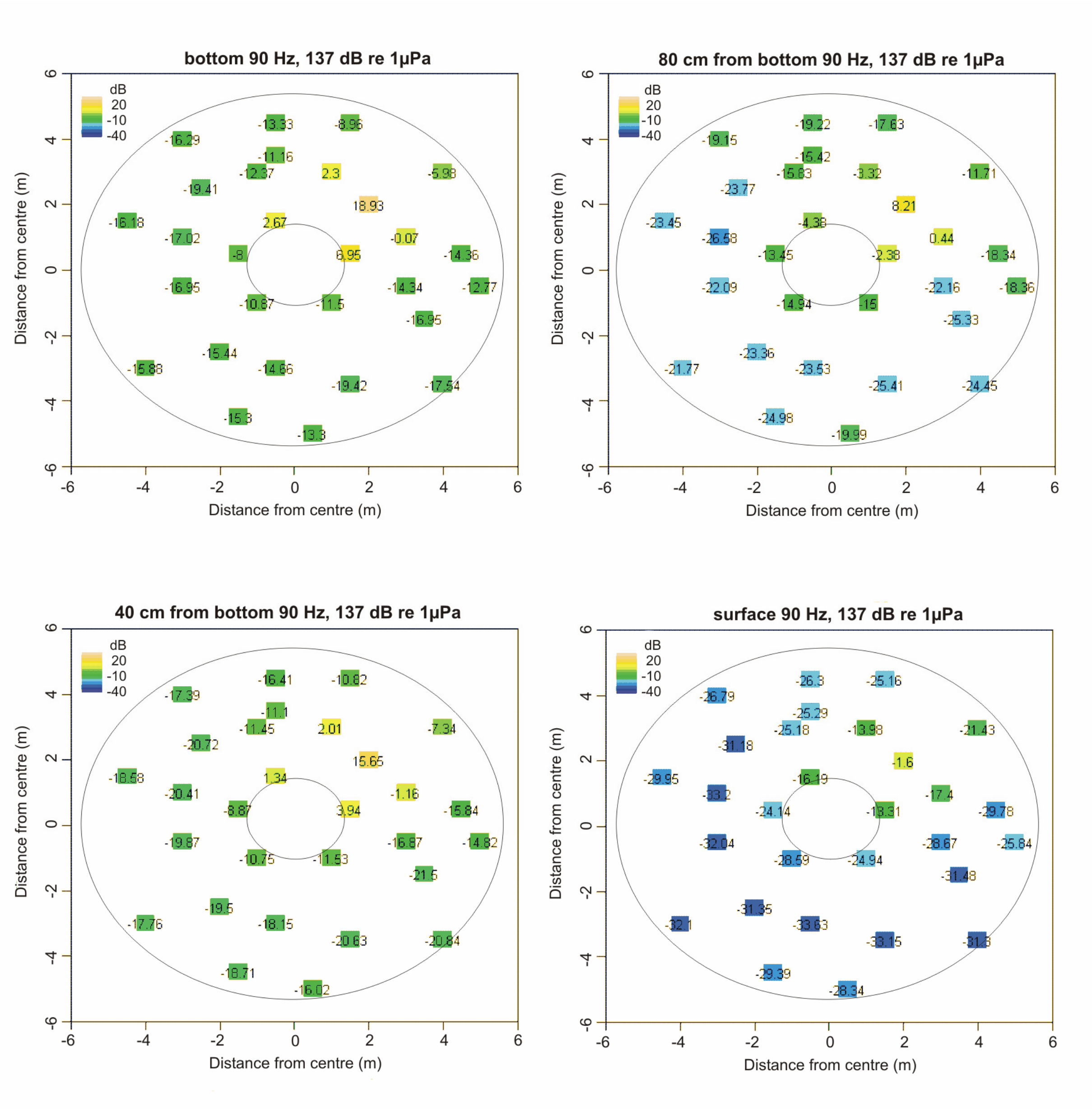

Fig. A 21: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 90 Hz and a sound level of 137 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

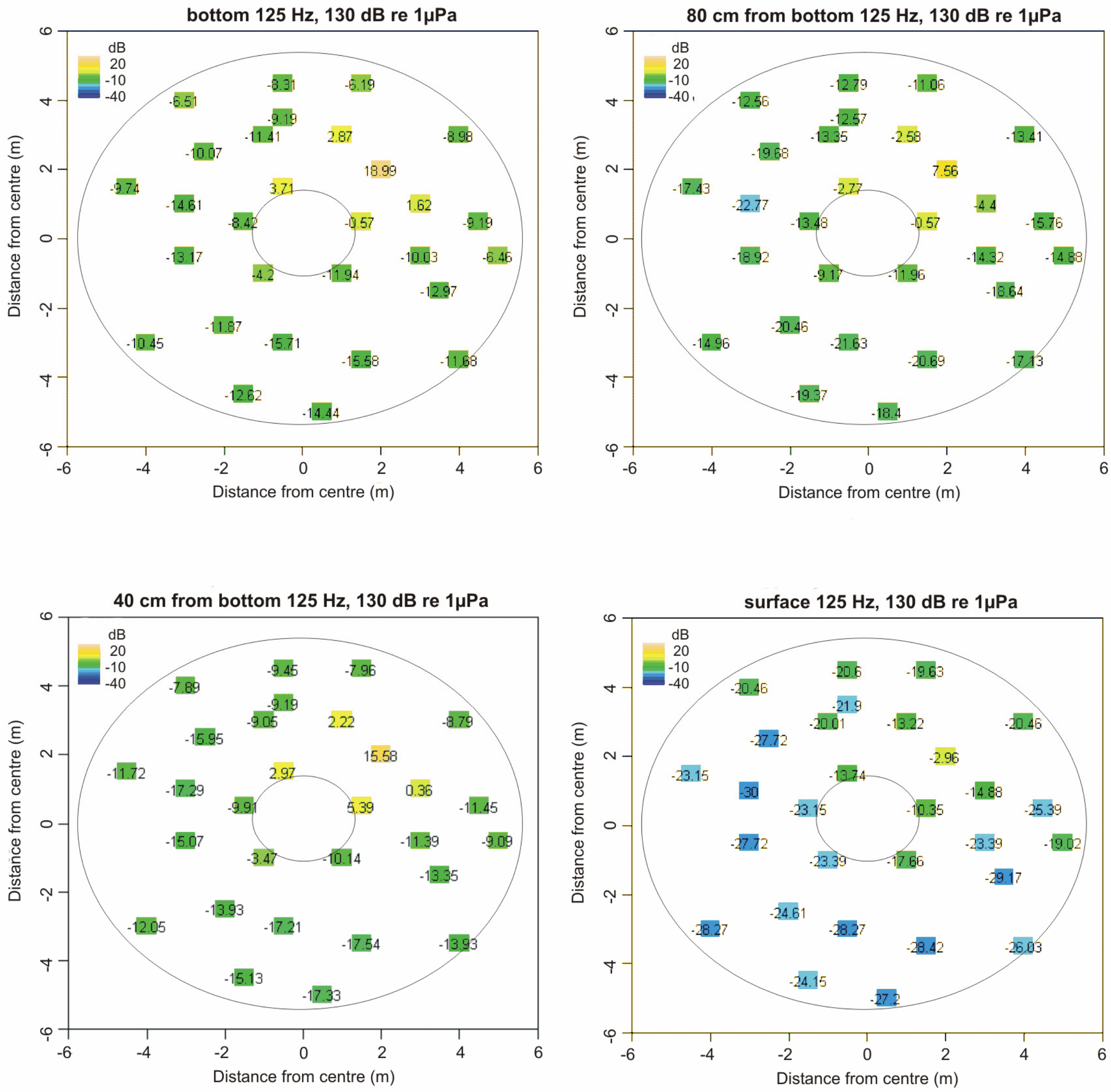

Fig. A 22: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 125 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

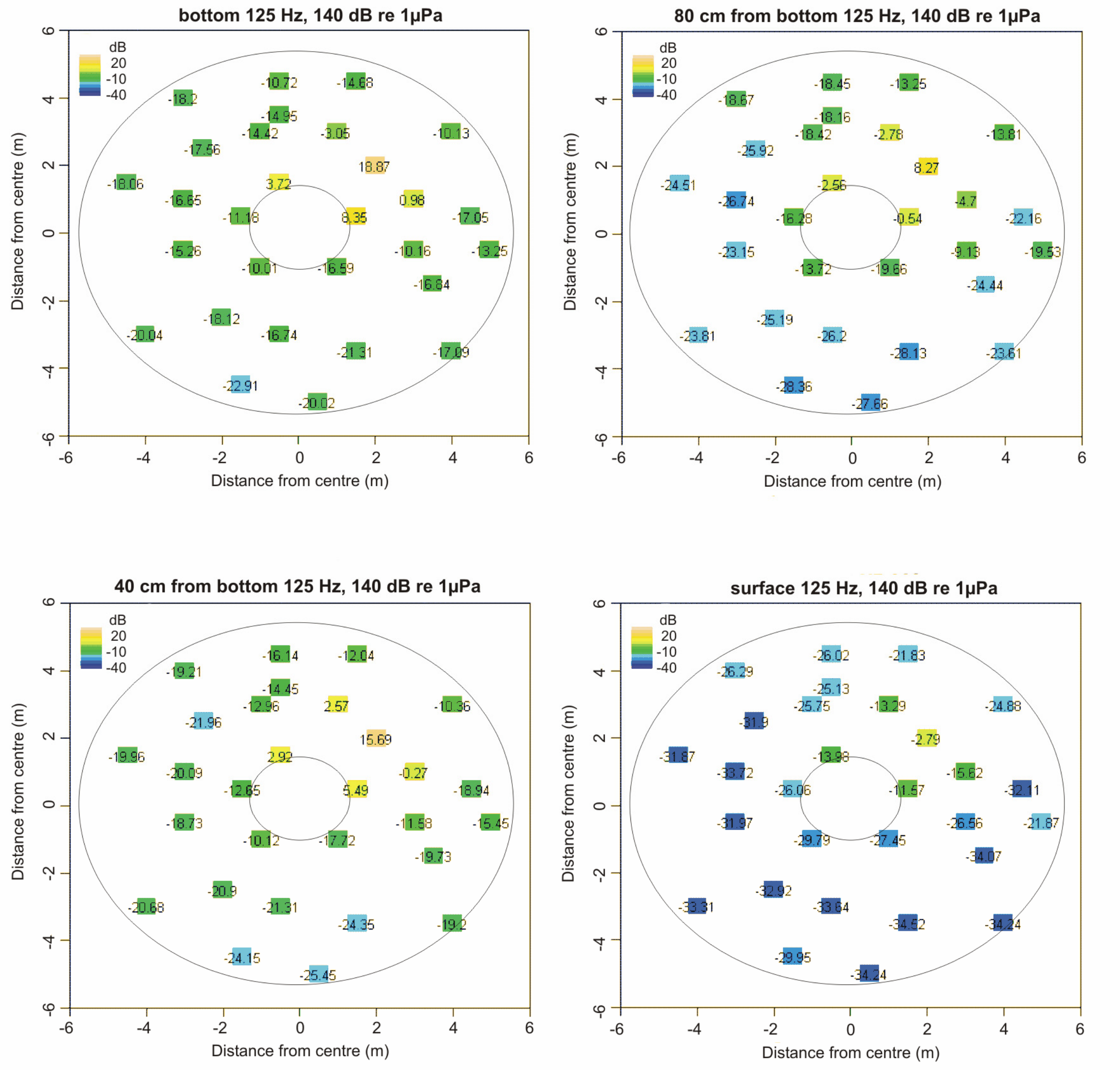

Fig. A 23: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 125 Hz and a sound level of 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

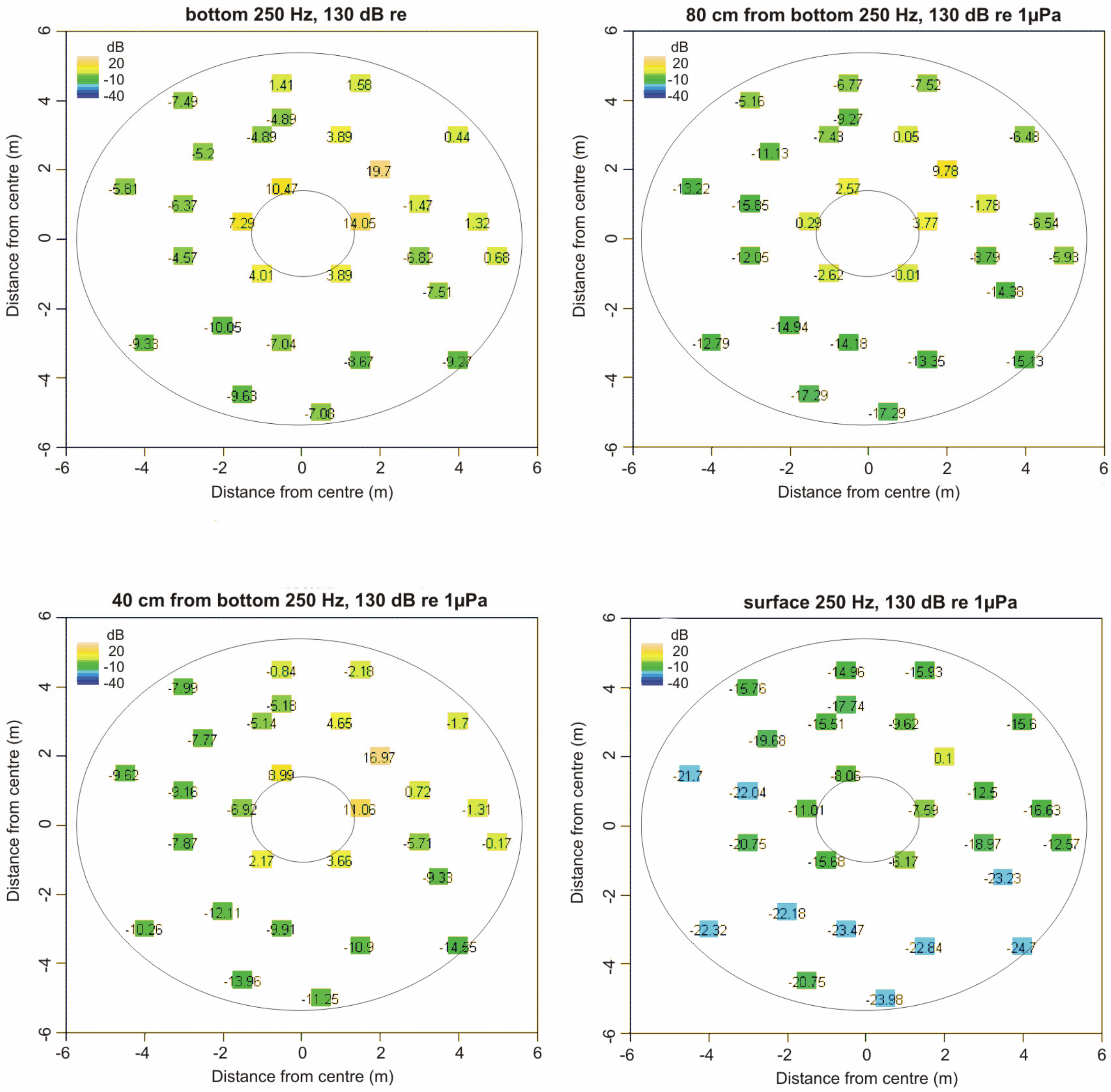

Fig. A 24: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 250 Hz and a sound level of 130 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

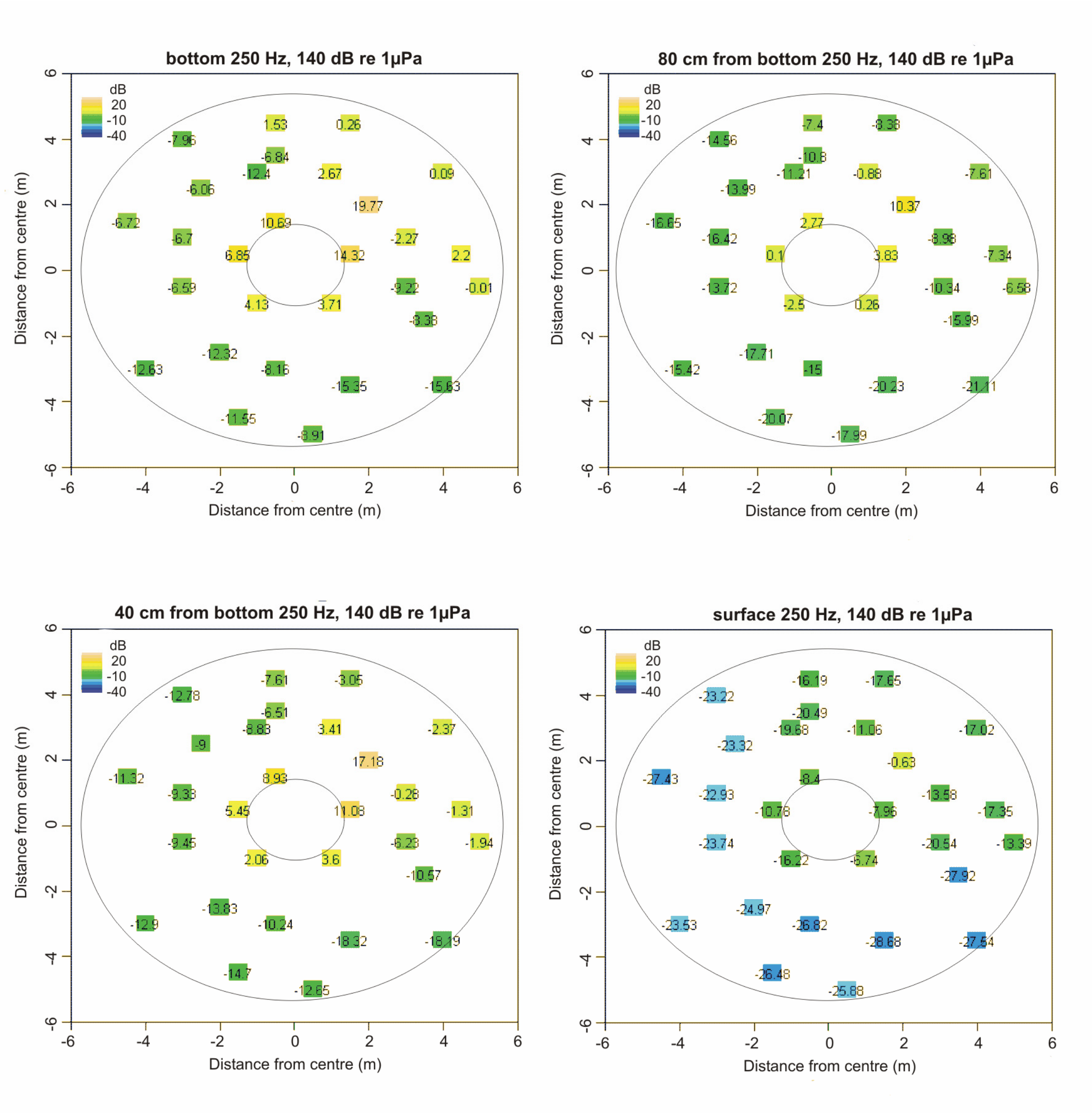

Fig. A 25: Sound field in the undivided tank during sound production in tank quarter 1 at a frequency of 250 Hz and a sound level of 140 dB re 1µPa. The measurements are presented as the difference from the produced sound level.

Table A 2: Sound pressure difference in the tank divided by barriers at different frequencies and sound levels calculated from minimal and maximal sound pressure levels at 86 measurement points.

25 Hz	Min	Max	Difference				
130 dB	[dB re 1µPa]		[dB]				
bottom	108,13	148,85	40,72				
40 cm from b.	105,34	145,58	40,24				
80 cm from b.	100,83	135,99	35,16				
surface	96,26	128,30	32,04				
60 Hz	Min	Max	Difference	**60 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	109,54	151,29	41,75	bottom	109,97	160,67	50,70
40 cm from b.	105,67	148,50	42,83	40 cm from b.	108,30	157,36	49,06
80 cm from b.	104,35	138,69	34,34	80 cm from b.	104,86	148,53	43,67
surface	97,50	129,43	31,93	surface	101,94	139,50	37,56
90 Hz	Min	Max	Difference	**90 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	110,24	151,34	41,10	bottom	113,62	160,59	46,97
40 cm from b.	106,85	148,50	41,65	40 cm from b.	107,23	158,51	51,28
80 cm from b.	103,52	138,28	34,76	80 cm from b.	103,52	149,25	45,73
surface	96,90	129,54	32,64	surface	98,59	140,36	41,77
125 Hz	Min	Max	Difference	**125 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	110,63	152,61	41,98	bottom	114,81	160,42	45,61
40 cm from b.	107,42	148,10	40,68	40 cm from b.	110,63	158,93	48,30
80 cm from b.	104,08	139,82	35,74	80 cm from b.	106,65	150,10	43,45
surface	98,28	130,88	32,60	surface	100,00	141,20	41,20
250 Hz	Min	Max	Difference	**250 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	106,85	154,82	47,97	bottom	111,25	160,59	49,34
40 cm from b.	106,65	151,41	44,76	40 cm from b.	108,94	160,59	51,65
80 cm from b.	103,23	141,66	38,43	80 cm from b.	103,81	153,03	49,22
surface	99,08	133,94	34,86	surface	100,00	143,94	43,94

Table A 3: Sound pressure difference in the tank divided by barriers at different frequencies and sound levels calculated from minimal and maximal sound pressure levels on the basis of 29 selected measurement points as used in the measurement in the undivided tank.

25 Hz	Min	Max	Difference				
130 dB	[dB re 1µPa]		[dB]				
bottom	108,13	148,85	40,72				
40 cm from b.	105,34	145,58	40,24				
80 cm from b.	100,83	135,56	34,73				
surface	98,06	127,00	28,94				
60 Hz	Min	Max	Difference	**60 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	109,54	150,66	41,12	bottom	109,97	160,67	50,70
40 cm from b.	105,80	147,53	41,73	40 cm from b.	108,30	157,36	49,06
80 cm from b.	104,61	138,17	33,56	80 cm from b.	104,86	147,64	42,78
surface	97,50	129,43	31,93	surface	101,94	139,50	37,56
90 Hz	Min	Max	Difference	**90 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	110,24	151,34	41,10	bottom	113,62	160,34	46,72
40 cm from b.	107,60	147,71	40,11	40 cm from b.	107,23	158,51	51,28
80 cm from b.	103,52	138,17	34,65	80 cm from b.	103,52	148,66	45,14
surface	98,59	129,54	30,95	surface	98,59	140,36	41,77
125 Hz	Min	Max	Difference	**125 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	110,63	151,69	41,06	bottom	114,89	160,34	45,45
40 cm from b.	107,42	148,10	40,68	40 cm from b.	110,63	158,93	48,30
80 cm from b.	104,08	138,89	34,81	80 cm from b.	108,03	149,31	41,28
surface	98,28	130,78	32,50	surface	101,58	141,20	39,62
250 Hz	Min	Max	Difference	**250 Hz**	Min	Max	Difference
130 dB	[dB re 1µPa]		[dB]	**140 dB**	[dB re 1µPa]		[dB]
bottom	106,85	154,25	47,40	bottom	111,25	160,00	48,75
40 cm from b.	106,65	150,42	43,77	40 cm from b.	108,94	160,59	51,65
80 cm from b.	103,23	141,66	38,43	80 cm from b.	104,86	151,91	47,05
surface	99,08	133,94	34,86	surface	100,42	143,94	43,52

Table A 4: Sound gradient in the undivided tank at different frequencies and sound levels calculated from minimal and maximal sound pressure levels at 29 measurement points.

25 Hz **121 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	107,16	137,09	29,93
40 cm from b.	103,97	133,16	29,19
80 cm from b.	101,29	125,58	24,29
surface	94,96	116,12	21,16

60 Hz **130 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	111,82	148,50	36,68
40 cm from b.	108,23	145,03	36,80
80 cm from b.	103,97	137,27	33,30
surface	98,06	125,11	27,05

90 Hz **130 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	113,80	148,91	35,11
40 cm from b.	112,04	145,55	33,51
80 cm from b.	107,89	138,01	30,12
surface	100,51	126,65	26,14

90 Hz **137 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	117,82	155,78	37,96
40 cm from b.	115,65	152,32	36,67
80 cm from b.	110,53	145,06	34,53
surface	103,52	134,89	31,37

125 Hz **130 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	114,29	148,99	34,70
40 cm from b.	112,46	145,58	33,12
80 cm from b.	107,23	137,56	30,33
surface	100,00	127,04	27,04

125 Hz **140 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	117,09	158,87	41,78
40 cm from b.	114,55	155,69	41,14
80 cm from b.	111,64	148,27	36,63
surface	105,48	137,21	31,73

250 Hz **130 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	119,95	149,70	29,75
40 cm from b.	115,45	146,97	31,52
80 cm from b.	112,71	139,78	27,07
surface	105,30	130,10	24,80

250 Hz **140 dB**	Min [dB re 1µPa]	Max	Difference [dB]
bottom	124,37	159,77	35,40
40 cm from b.	121,68	157,18	35,50
80 cm from b.	118,89	150,37	31,48
surface	111,32	139,37	28,05

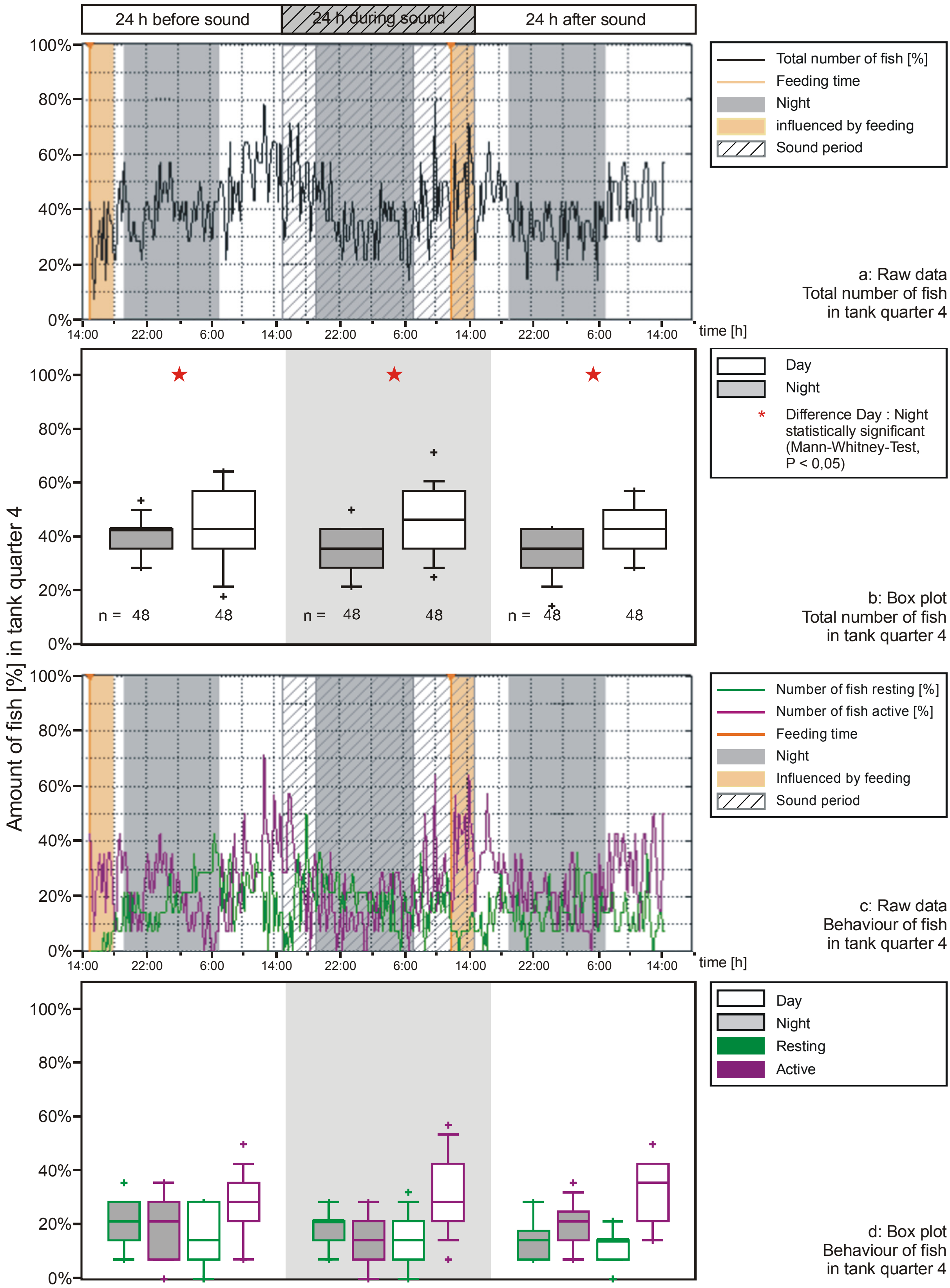

Fig. A 26: Results of sound experiment: Juvenile cod, 25 Hz, 130 dB re 1µPa.

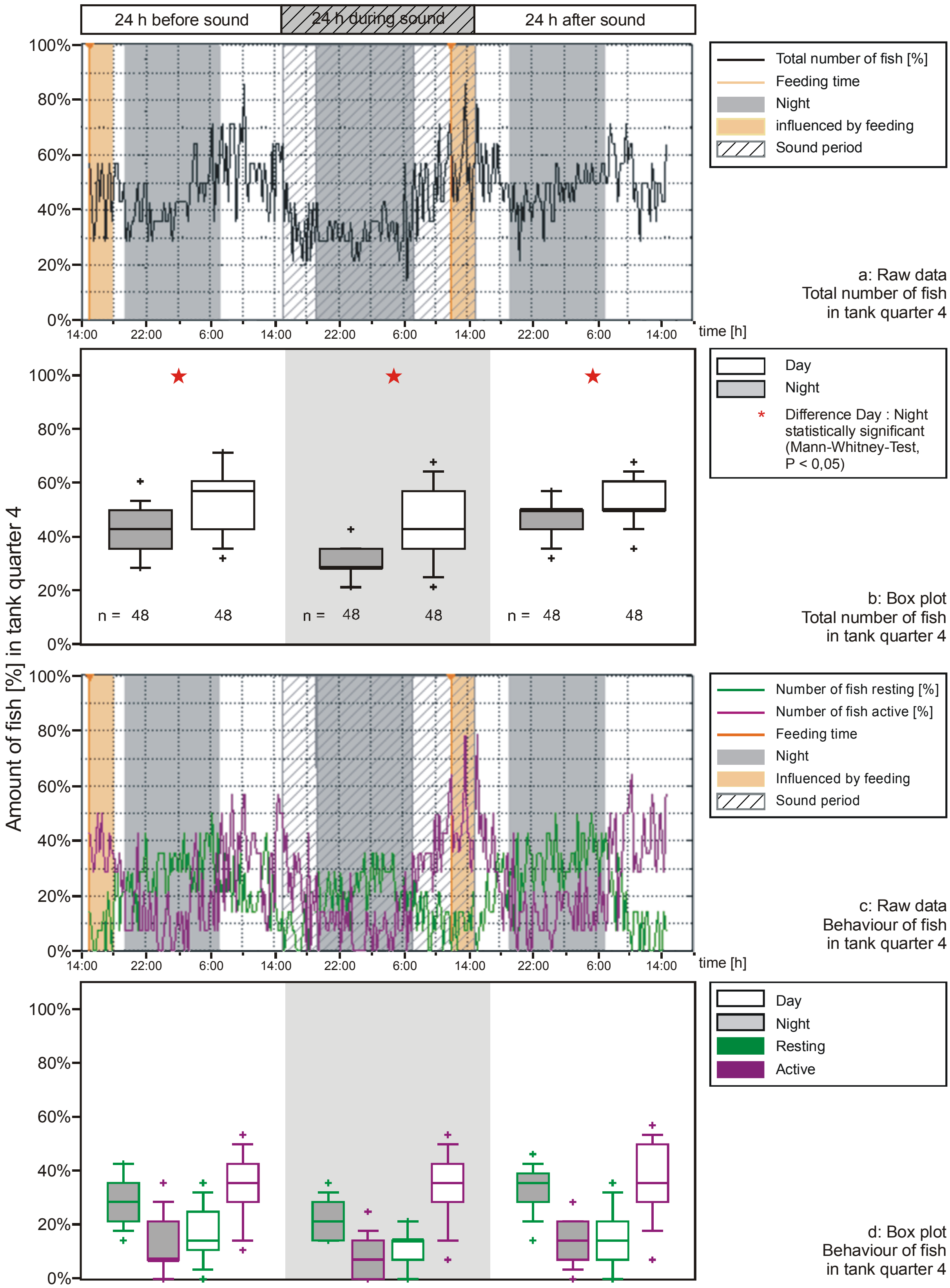

Fig. A 27: Results of sound experiment: Juvenile cod, 25 Hz, 140 dB re 1µPa.

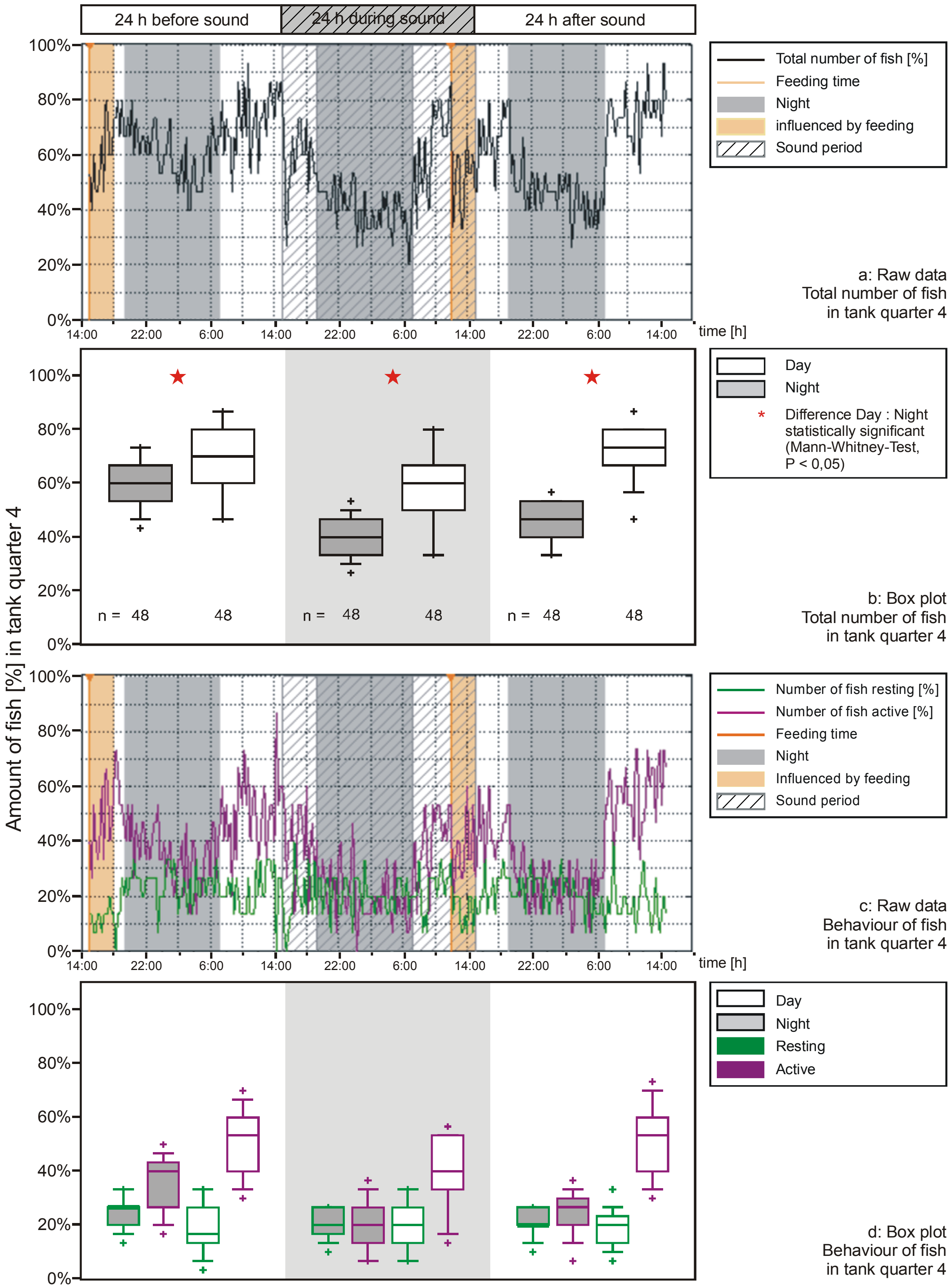

Fig. A 28: Results of sound experiment: Juvenile cod, 60 Hz, 130 dB re 1µPa.

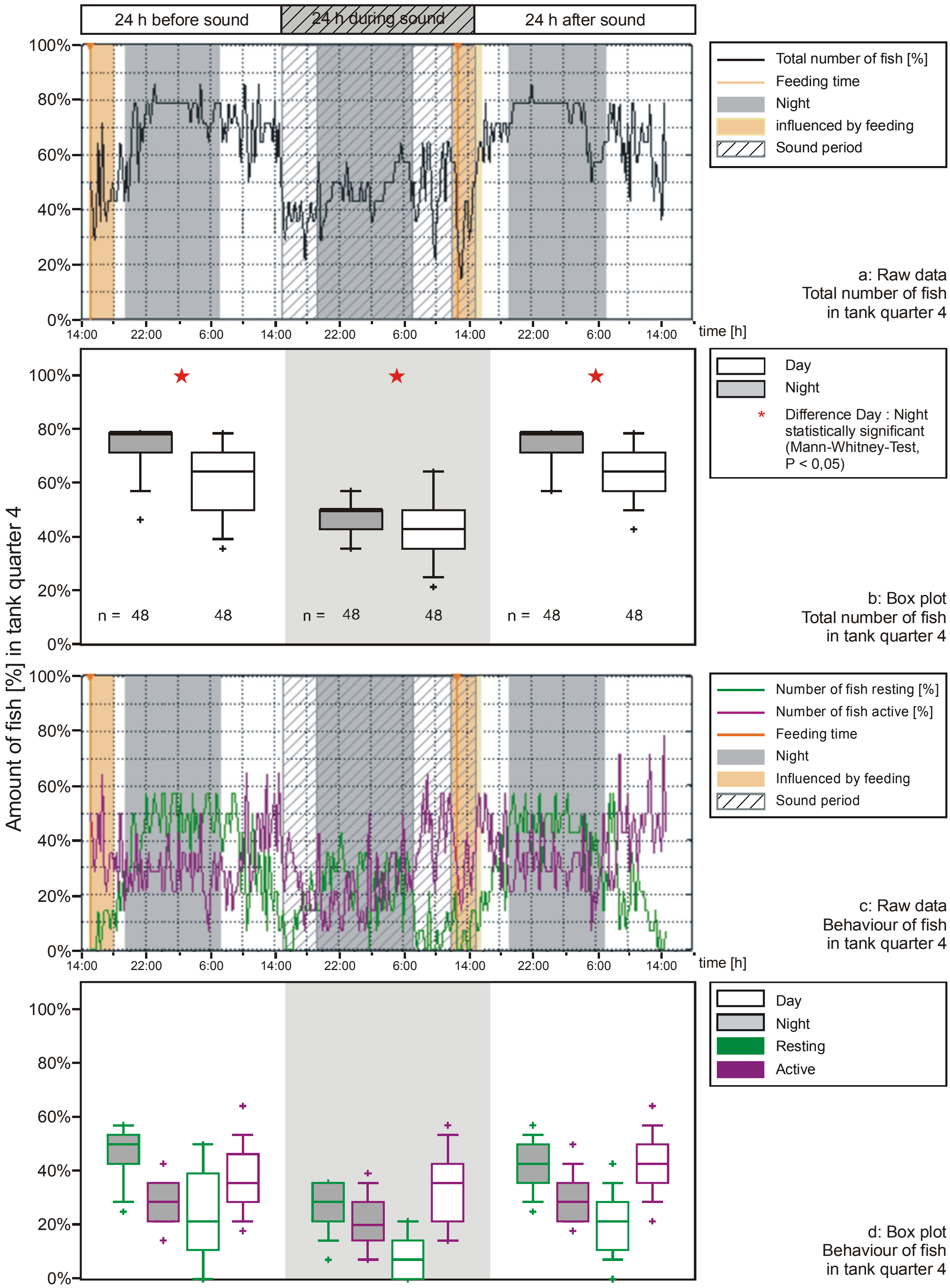

Fig. A 29: Results of sound experiment: Juvenile cod, 60 Hz, 140 dB re 1µPa.

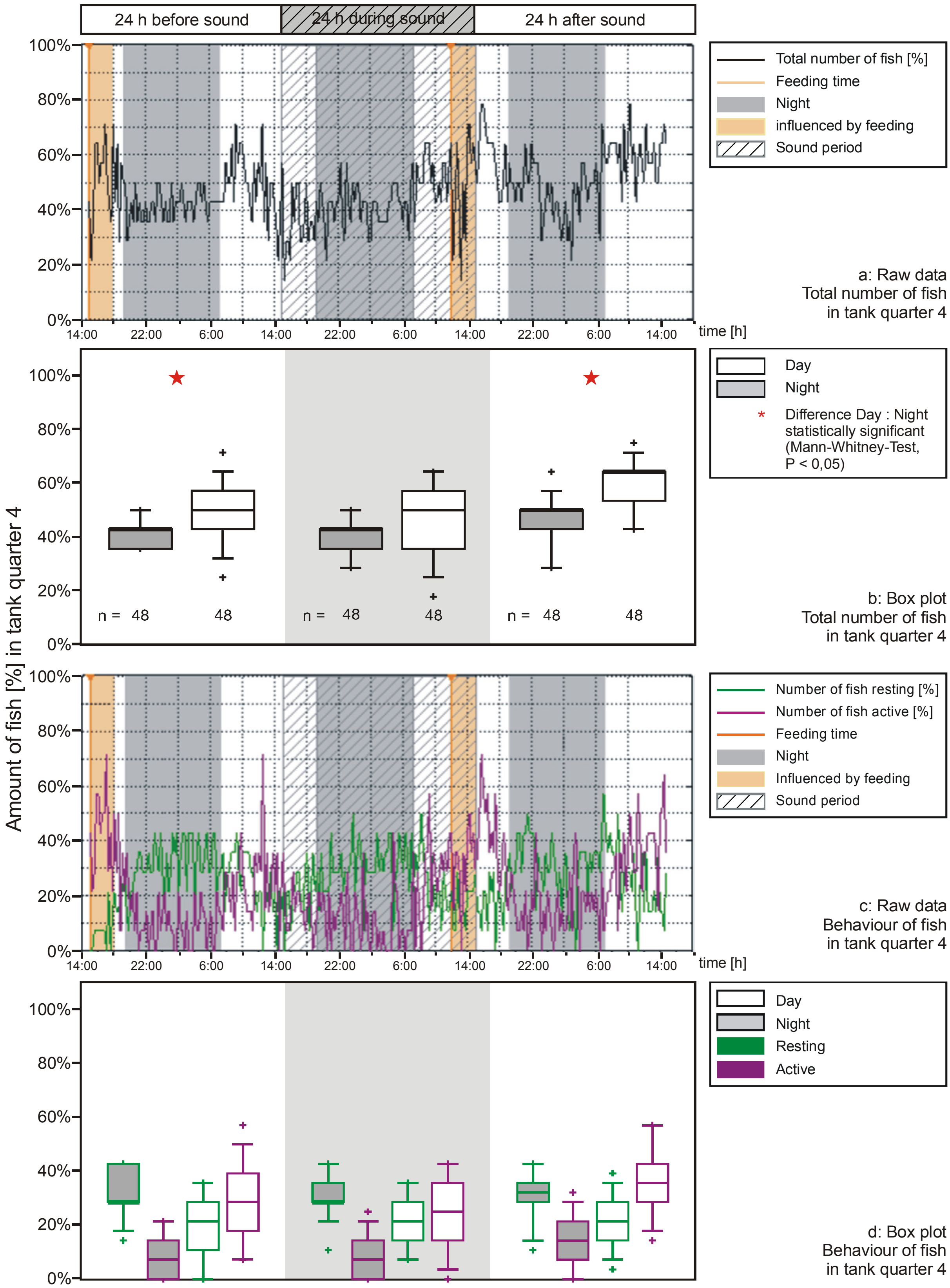

Fig. A 30: Results of sound experiment: Juvenile cod, 90 Hz, 130 dB re 1μPa.

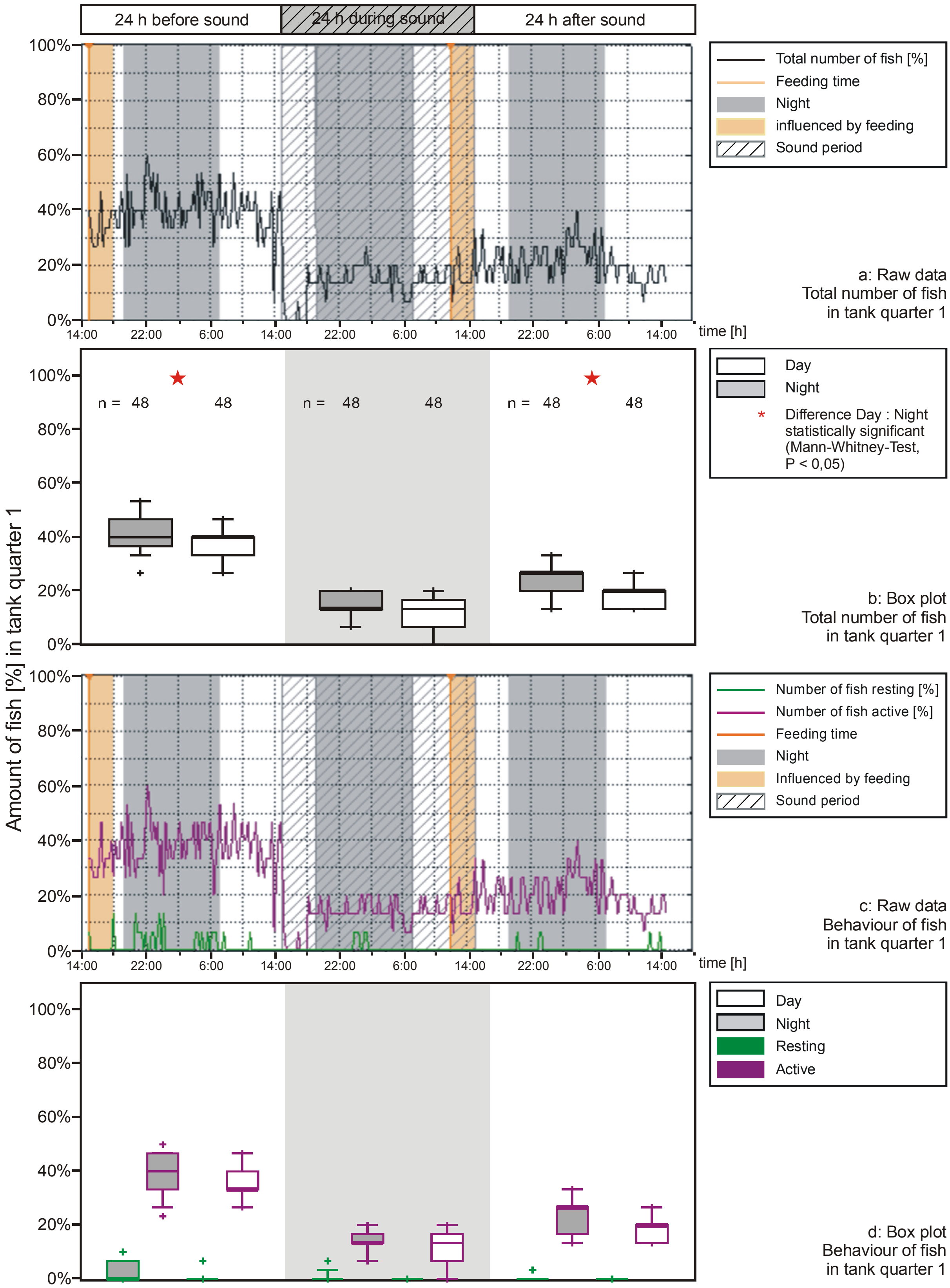

Fig. A 31: Results of sound experiment: Juvenile cod, 90 Hz, 140 dB re 1µPa. Sound production in tank quarter 1

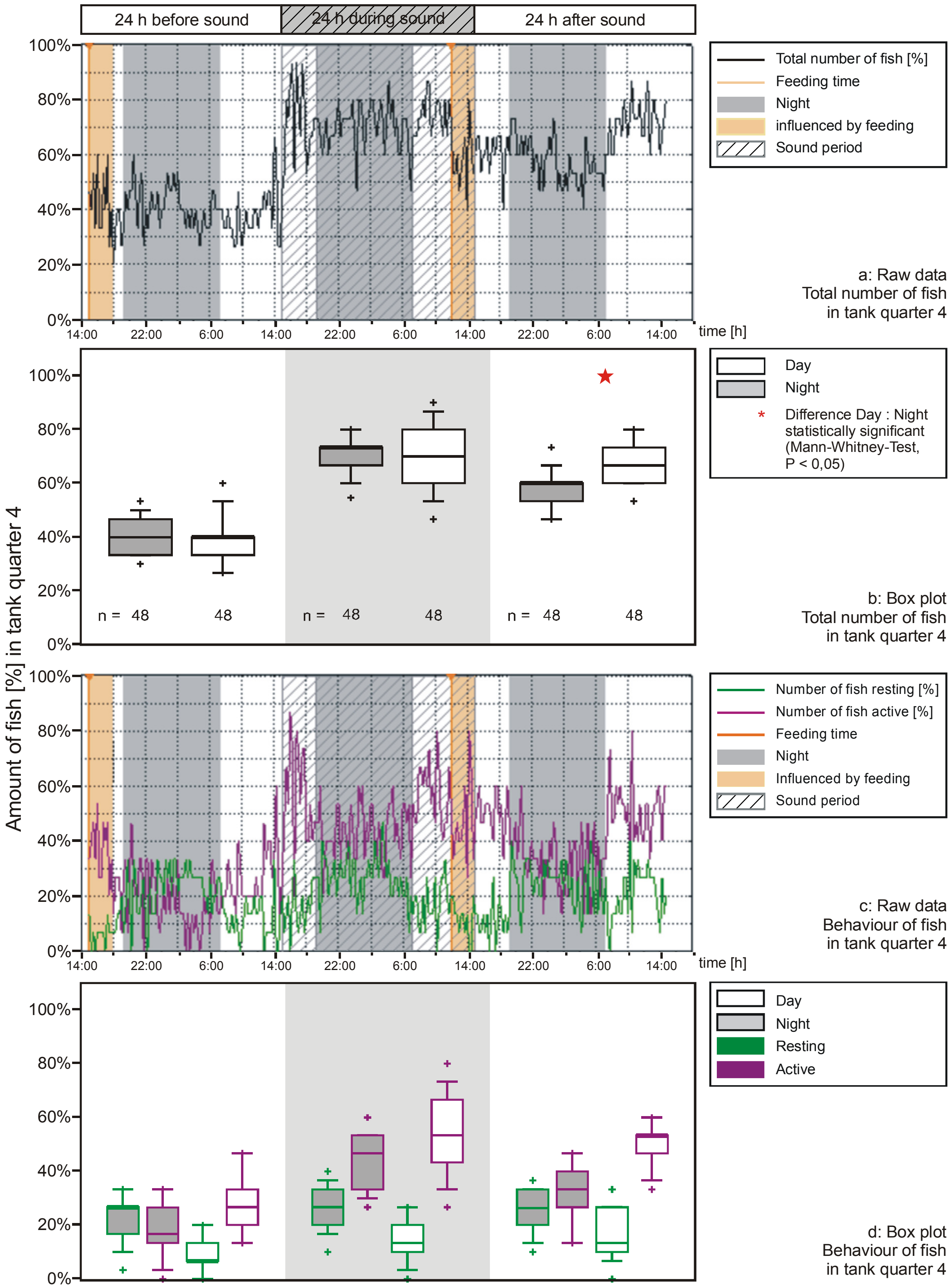

Fig. A 32: Results of sound experiment: Juvenile cod, 90 Hz, 140 dB re 1µPa. Sound production in tank quarter 1.

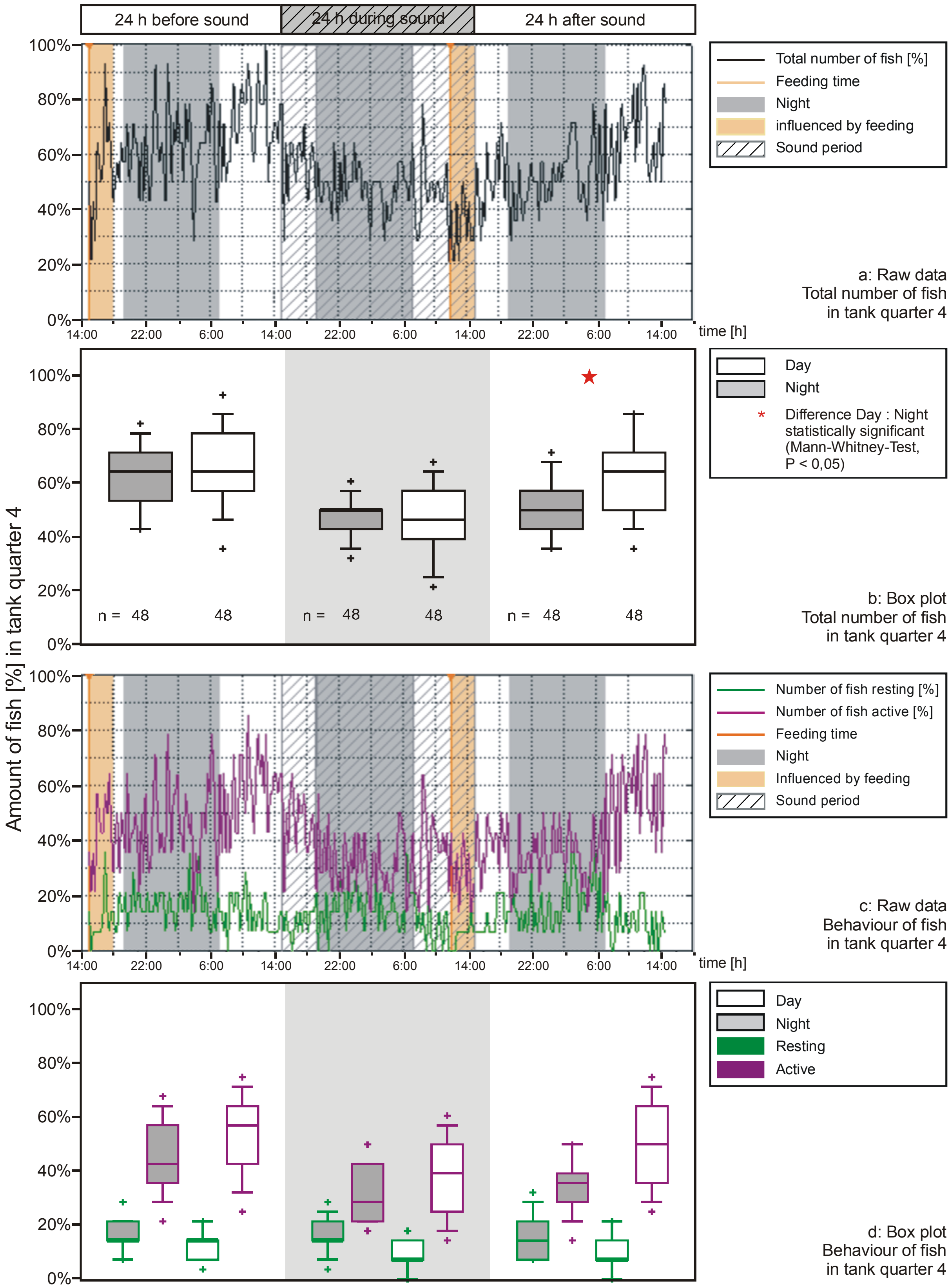

Fig. A 33: Results of sound experiment: Juvenile cod, 125 Hz, 130 dB re 1µPa.

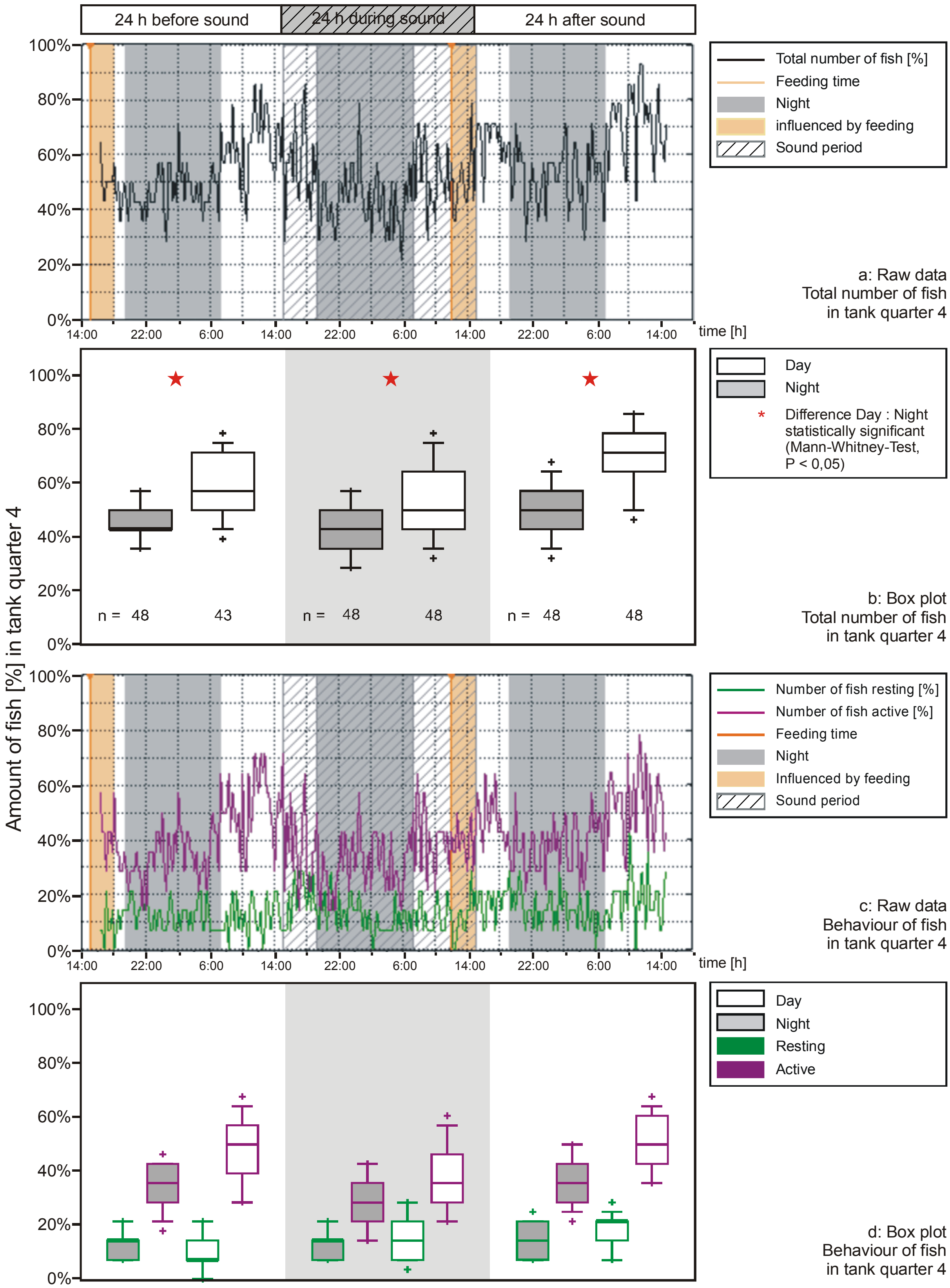

Fig. A 34: Results of sound experiment: Juvenile cod, 125 Hz, 140 dB re 1µPa.

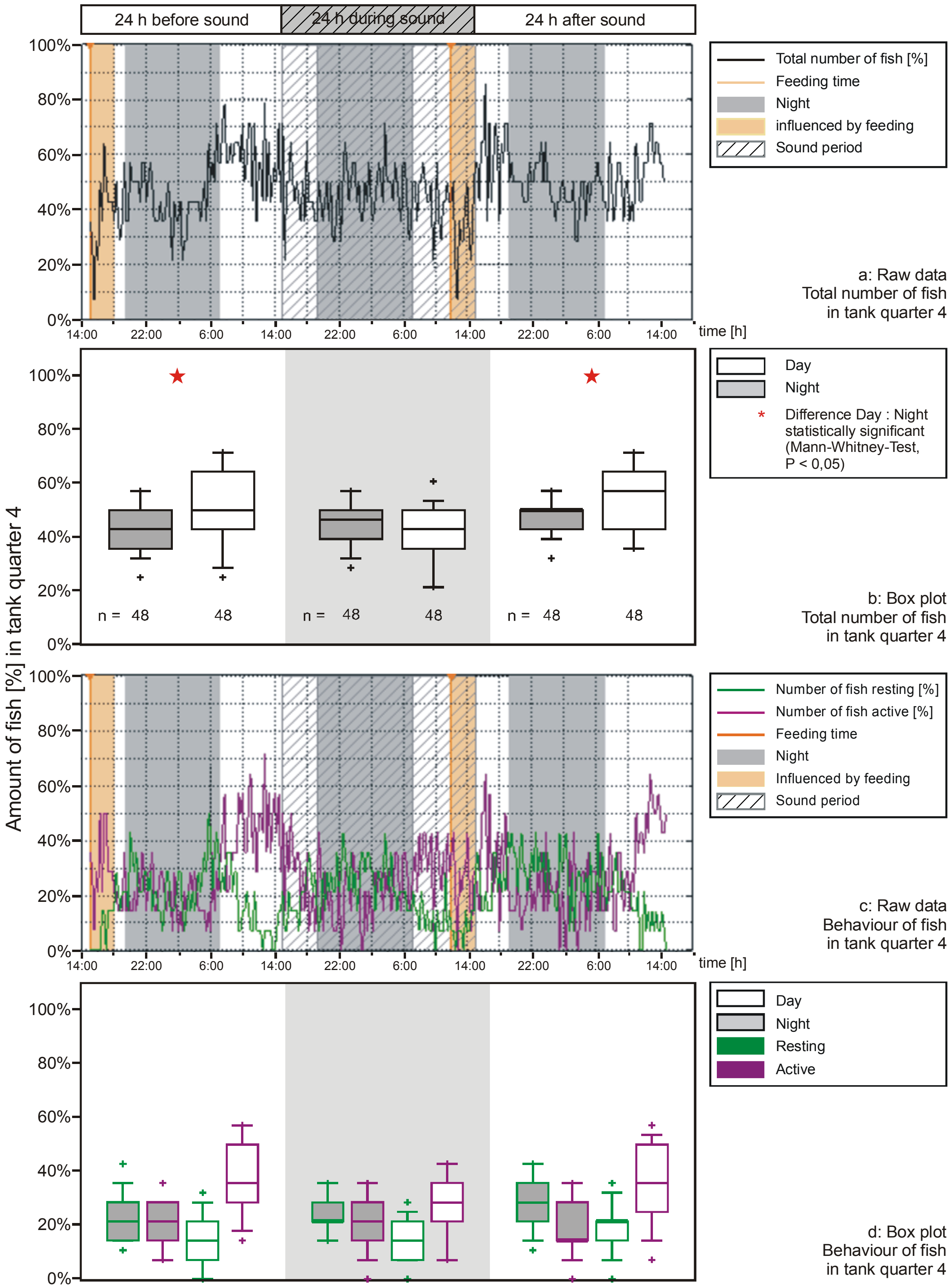

Fig. A 35: Results of sound experiment: Juvenile cod, 250 Hz, 140 dB re 1µPa.

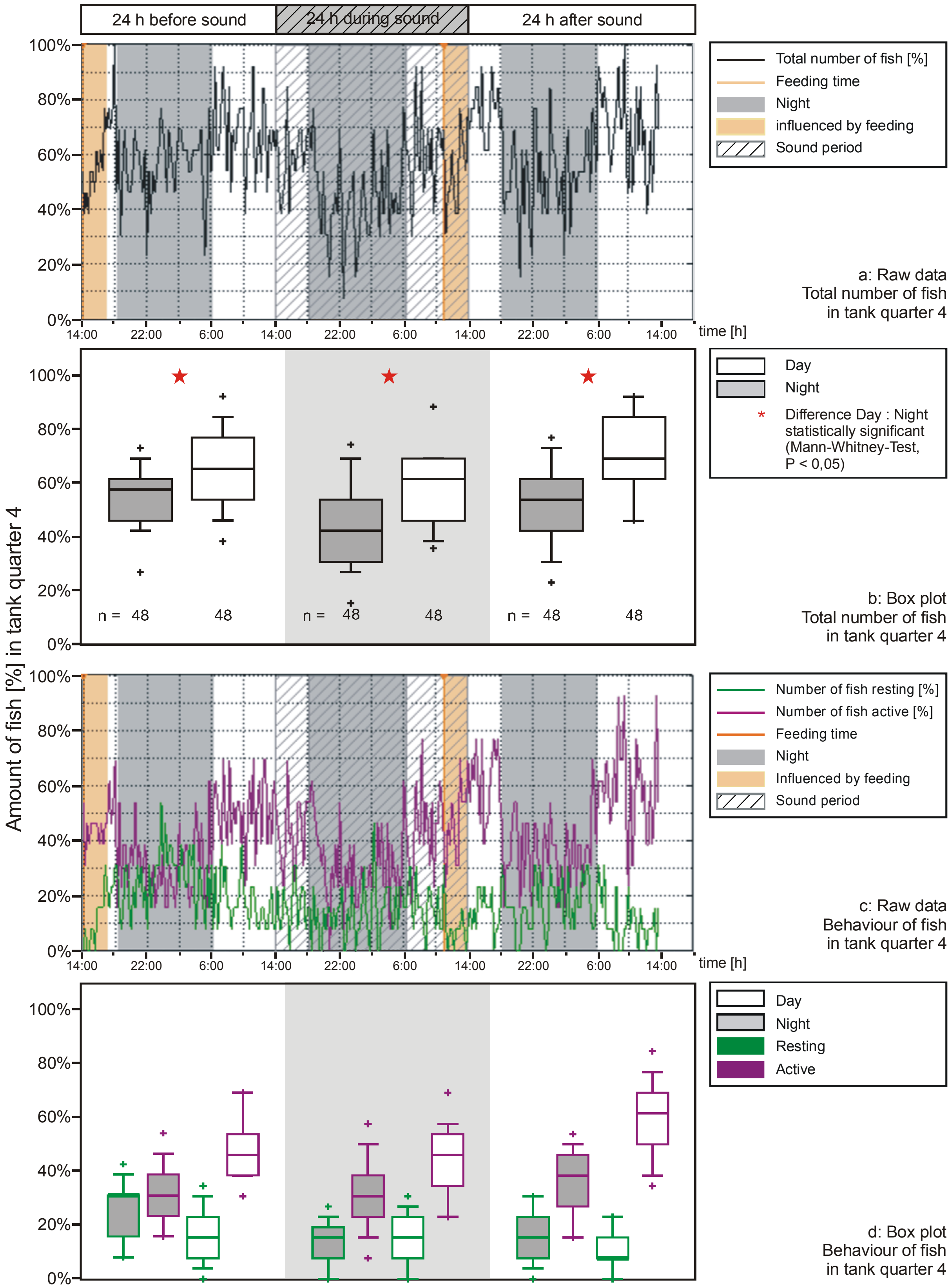

Fig. A 36: Results of sound experiment: Adult cod, 25 Hz, 130 dB re 1μPa.

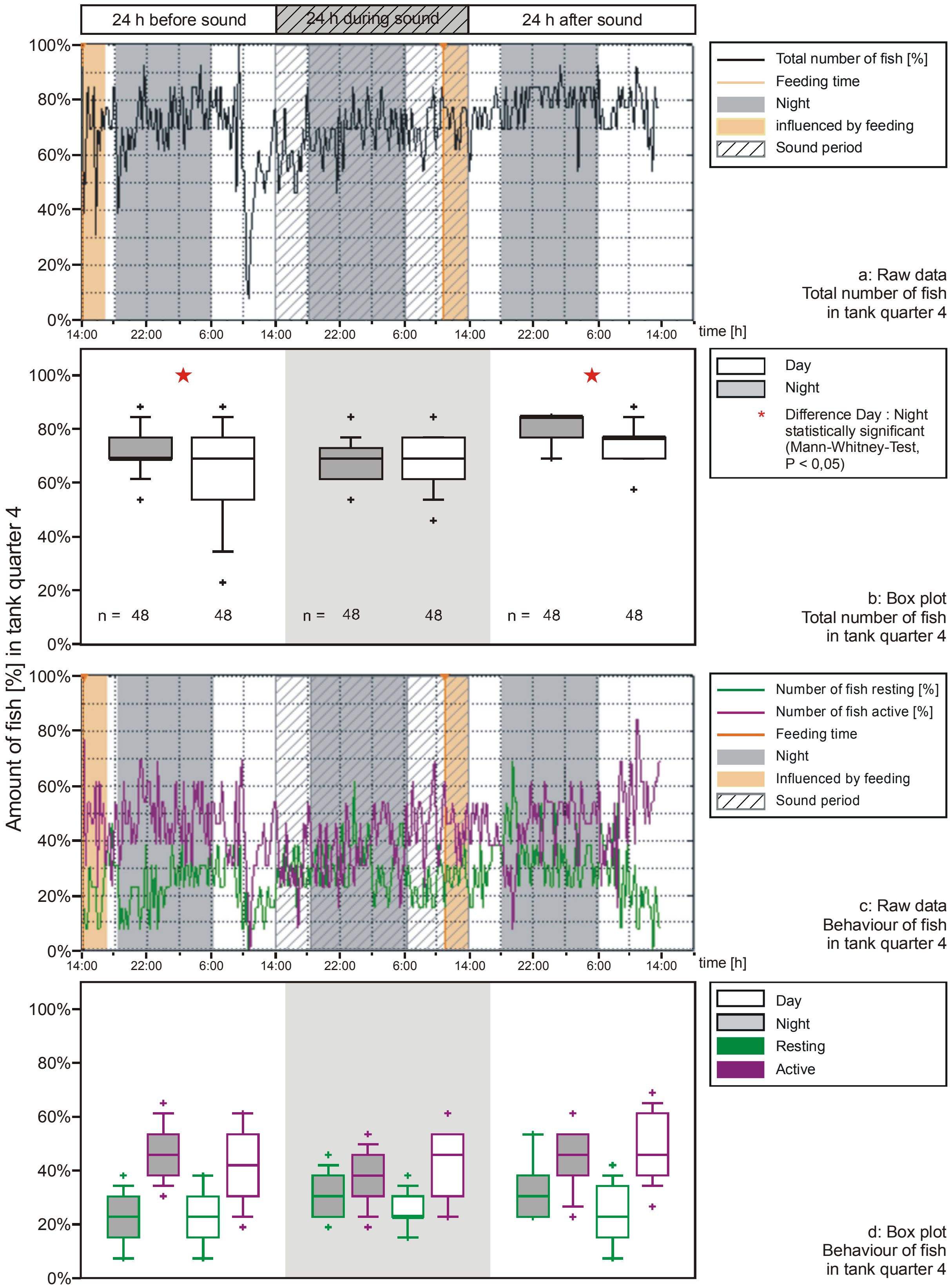

Fig. A 37: Results of sound experiment: Adult cod, 25 Hz, 140 dB re 1μPa.

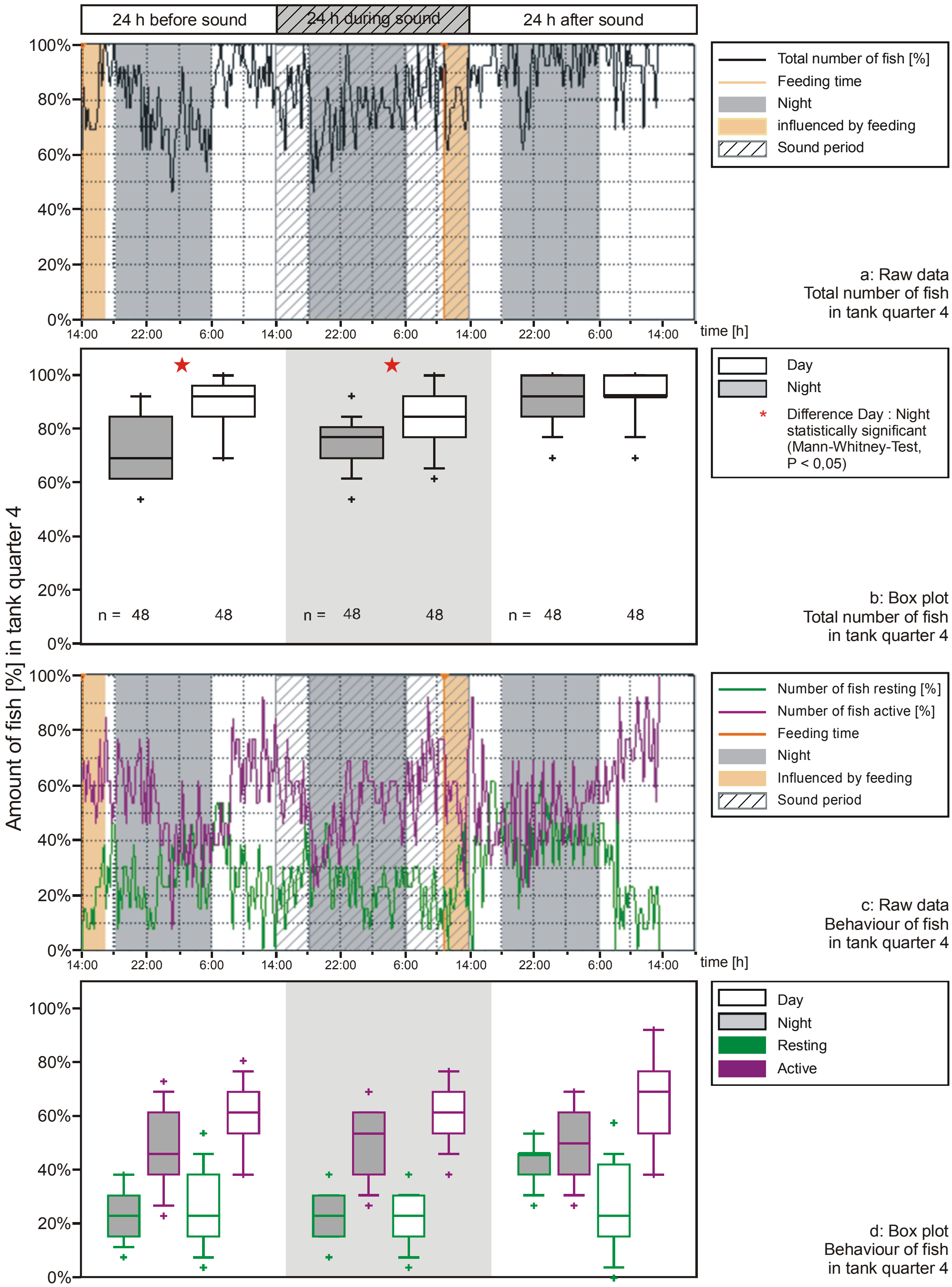

Fig. A 38: Results of sound experiment: Adult cod, 60 Hz, 130 dB re 1µPa.

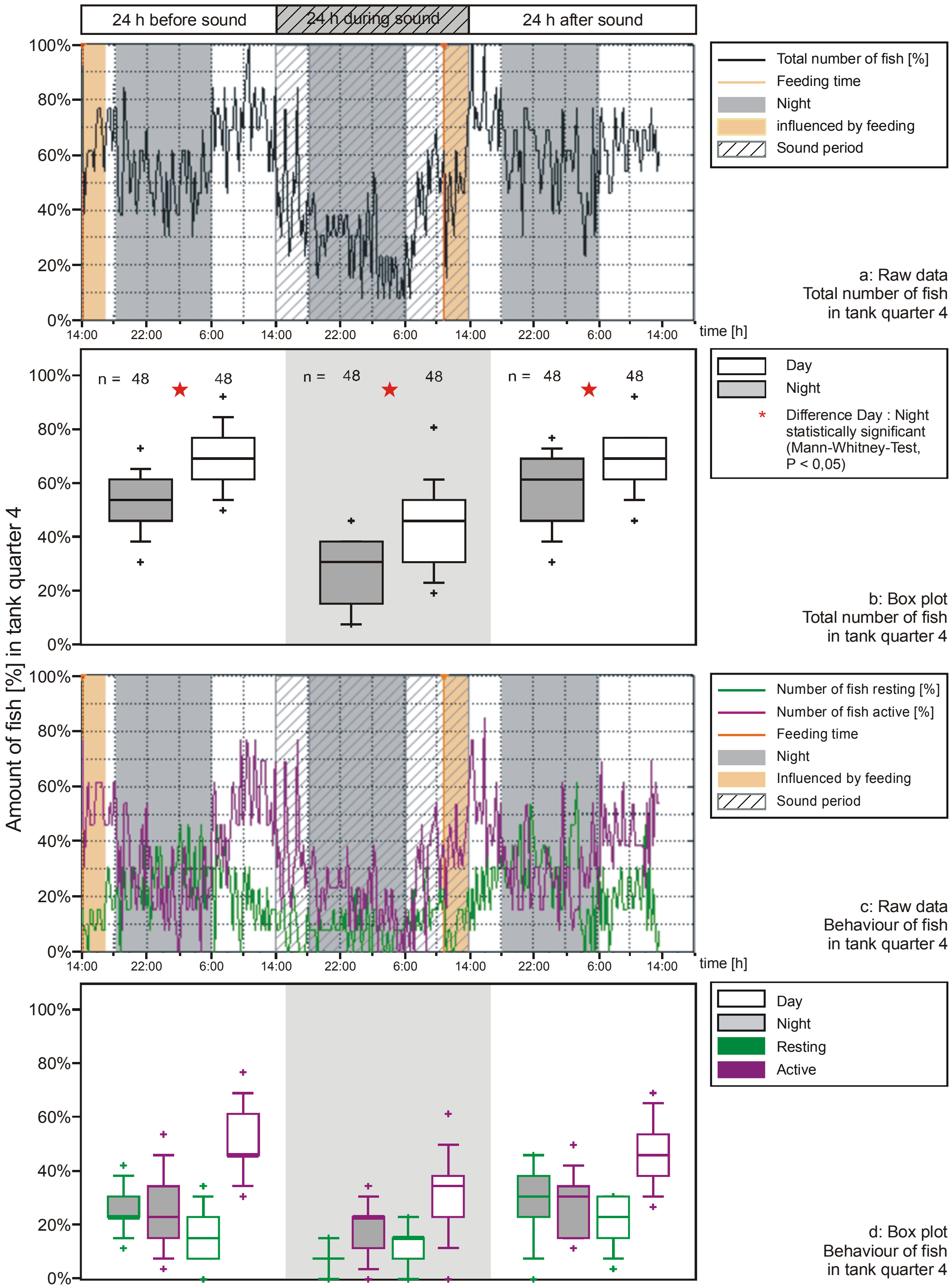

Fig. A 39: Results of sound experiment: Adult cod, 60 Hz, 140 dB re 1µPa.

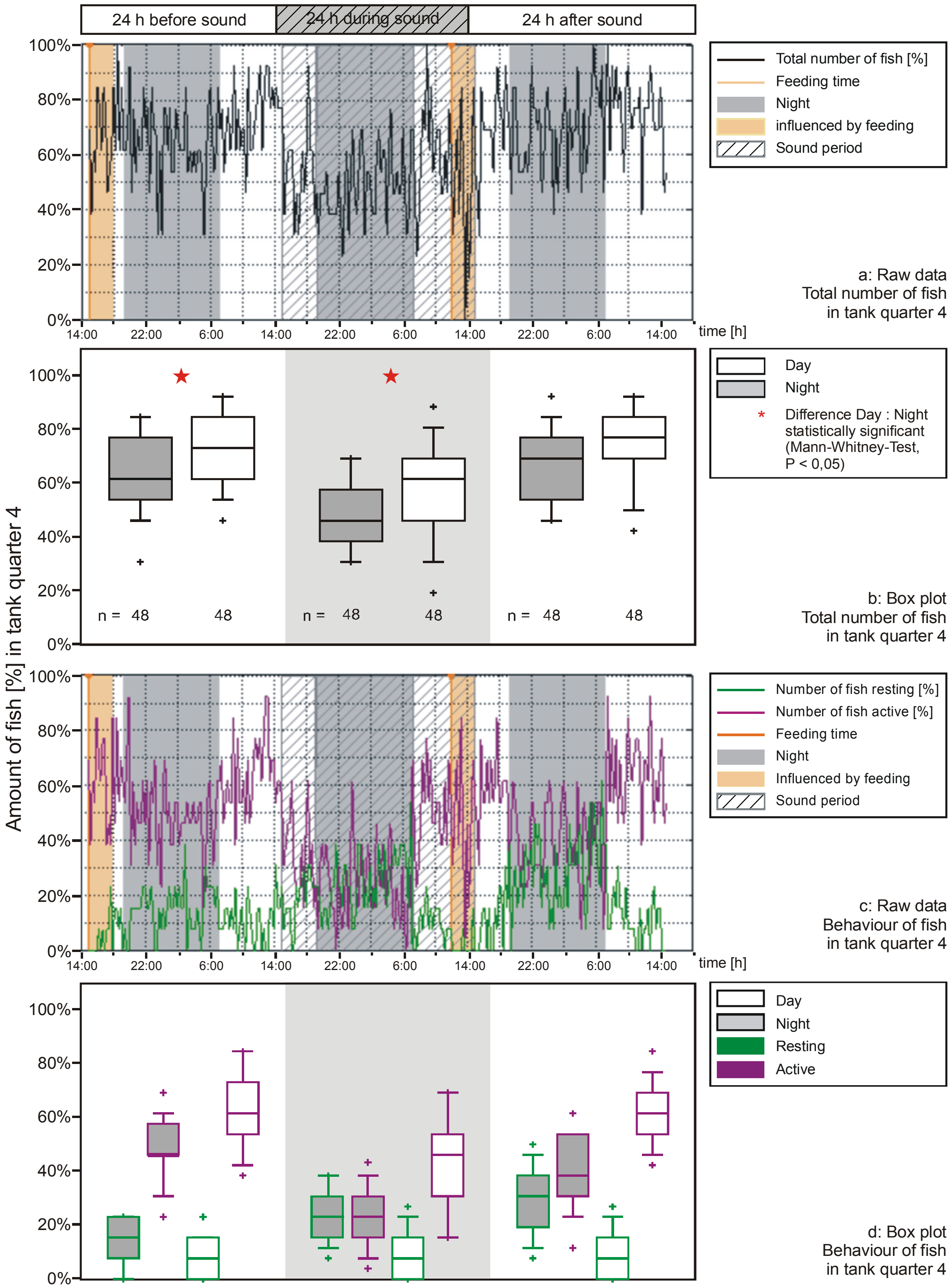

Fig. A 40: Results of sound experiment: Adult cod, 90 Hz, 130 dB re 1µPa.

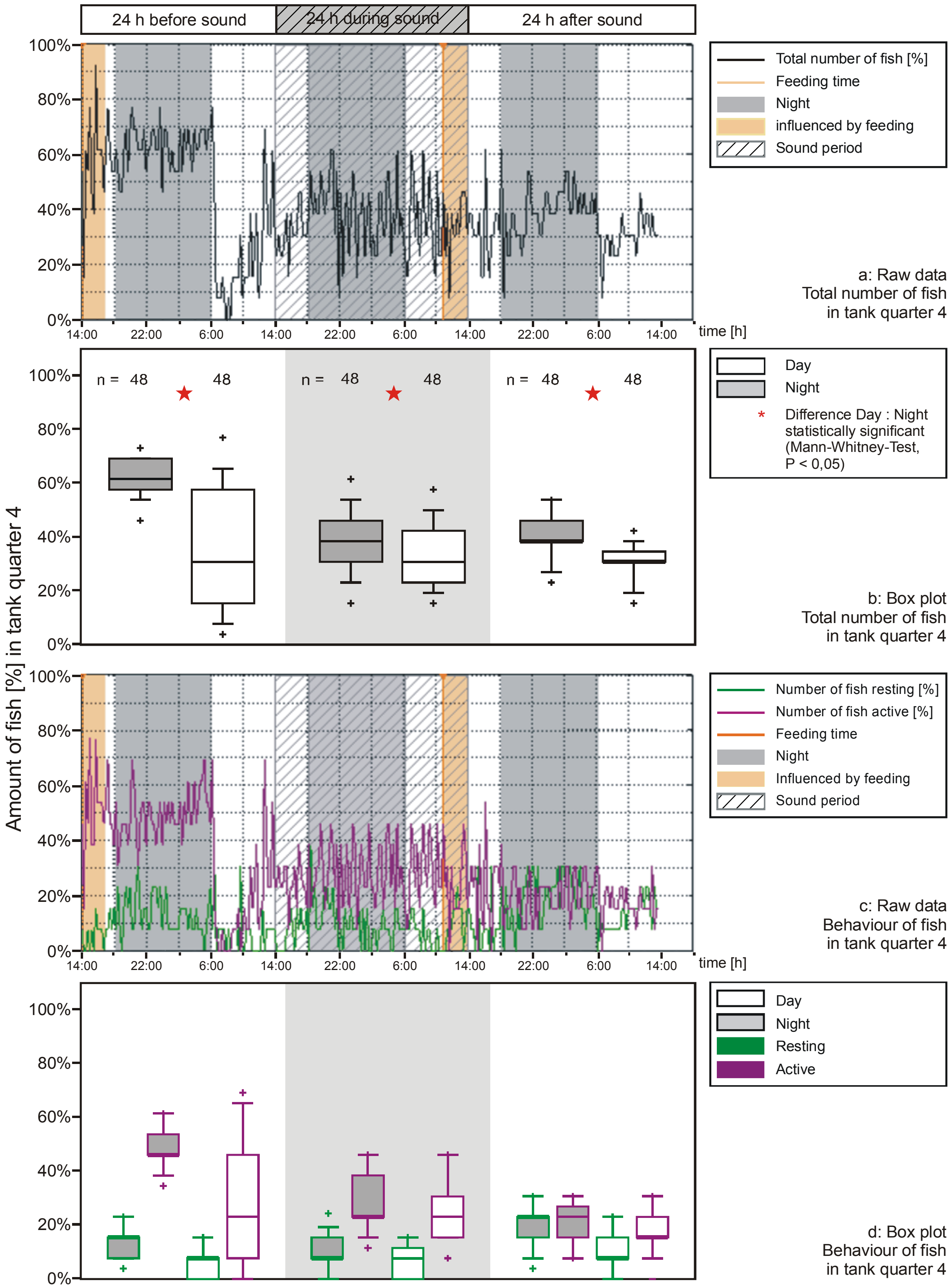

Fig. A 41: Results of sound experiment: Adult cod, 90 Hz, 140 dB re 1µPa.

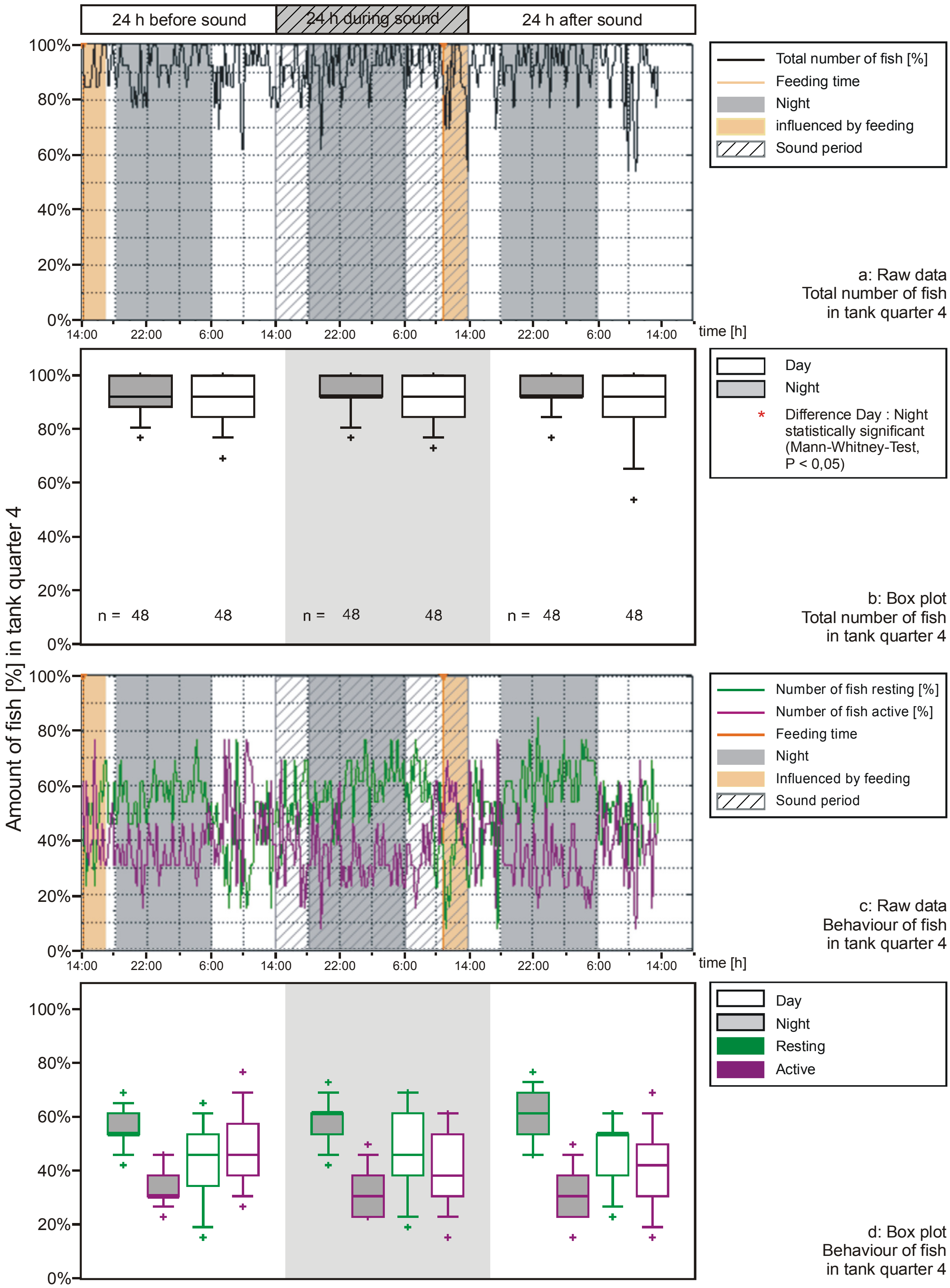

Fig. A 42: Results of sound experiment: Adult cod, 125 Hz, 130 dB re 1µPa.

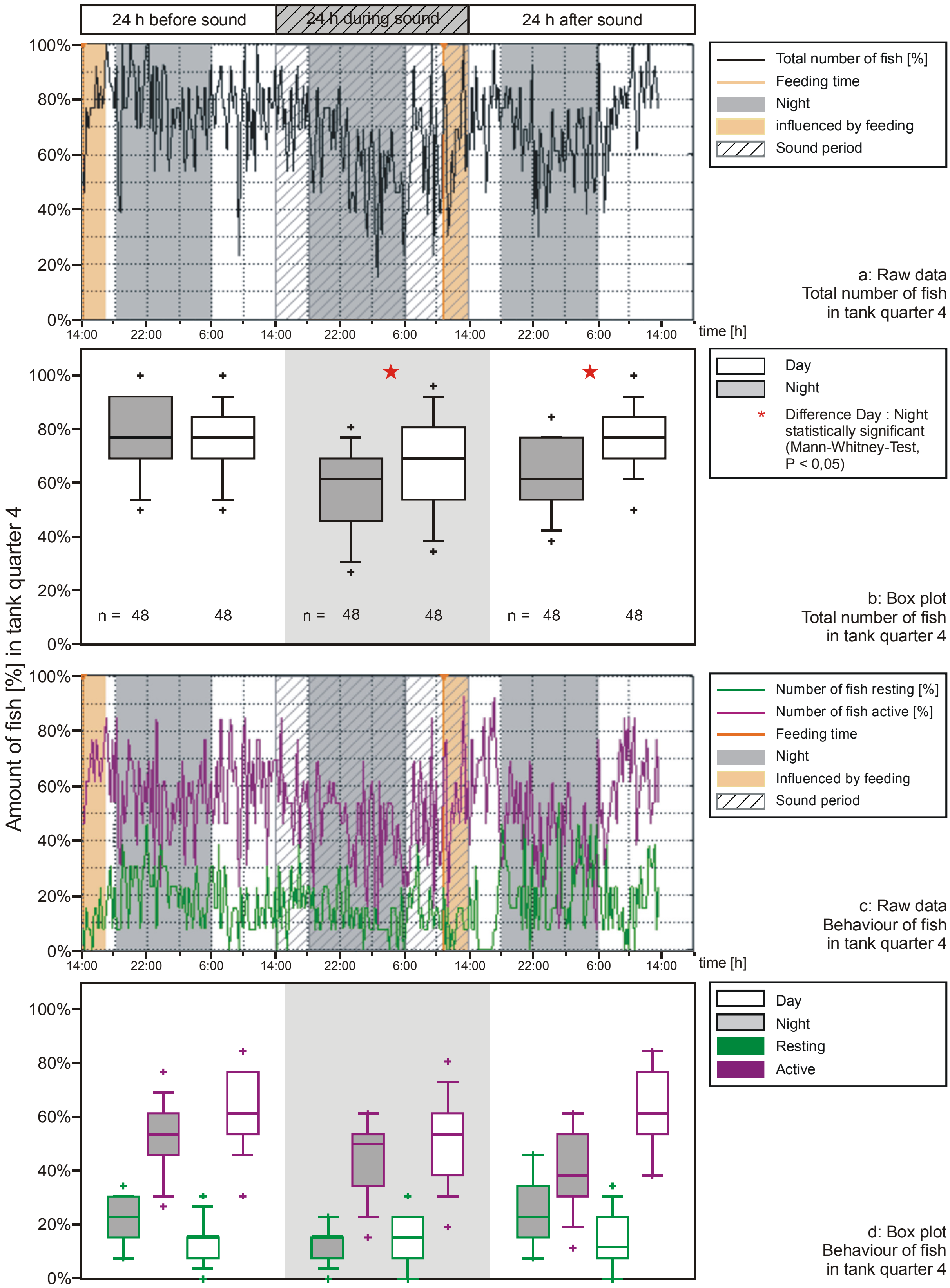

Fig. A 43: Results of sound experiment: Adult cod, 125 Hz, 140 dB re 1μPa.

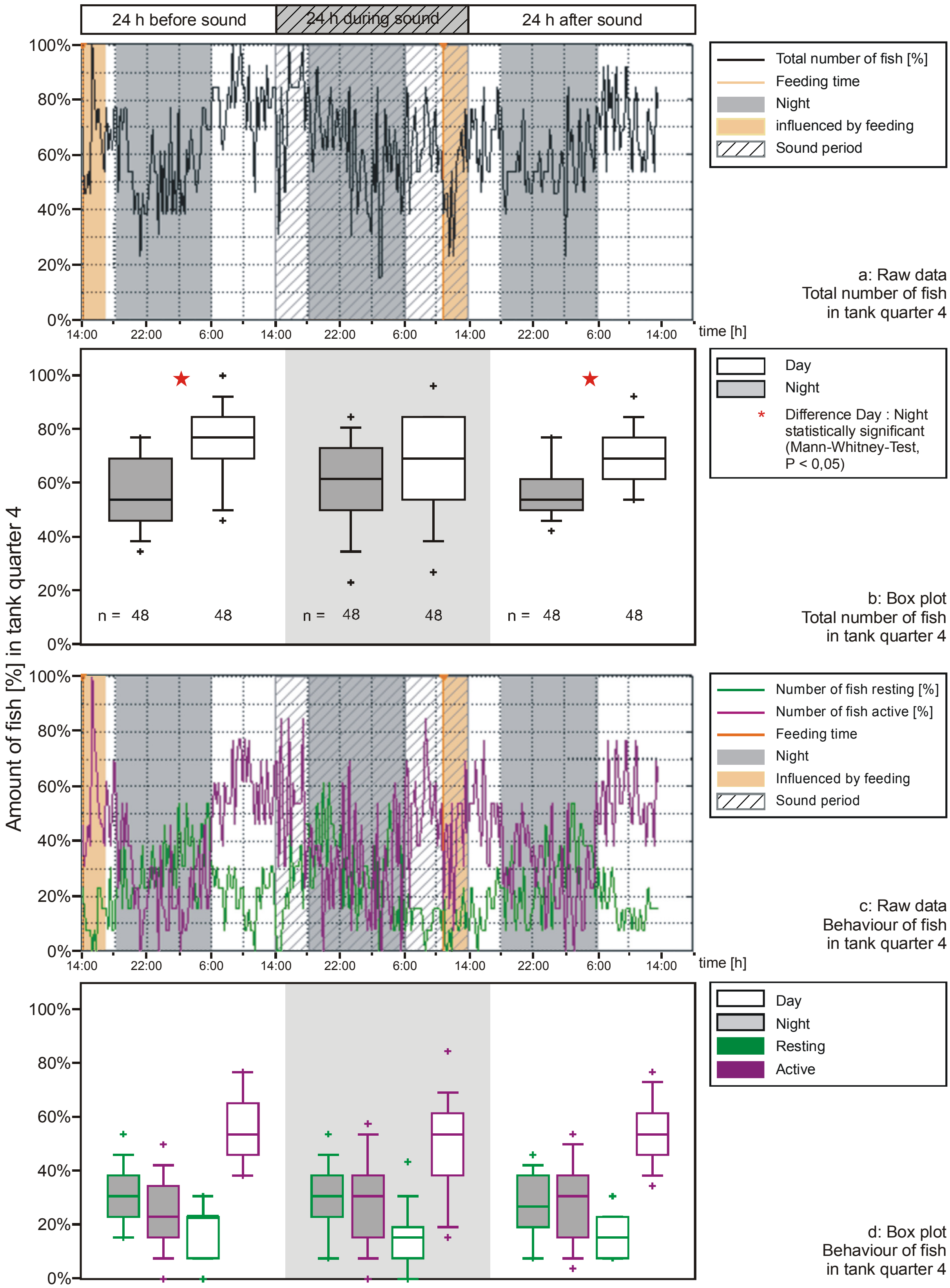

Fig. A 44: Results of sound experiment: Adult cod, 250 Hz, 140 dB re 1µPa.

Table A 5: Statistical results (Mann-Whitney U-Test) for significant differences in cod numbers in tank quarter 4 between day and night during the periods before, during and after sound production. Sound levels in dB re 1µPa.

Sound situation	before sound	during sound	after sound
	cod juvenile		
25 Hz/130 dB	0.0090	0.0000	0.0000
25 Hz/140 dB	0.0000	0.0000	0.0007
60 Hz/130 dB	0.0002	0.0000	0.0000
60 Hz/140 dB	0.0000	0.0037	0.0000
90 Hz/130 dB	0.0003	0.0239	0.0000
90 Hz/140 dB Quarter 1	0.0070	0.0572	0.0001
90 Hz/140 dB Quarter 4	0.0314	0.3819	0.0000
125 Hz/130 dB	0.1227	0.4230	0.0005
125 Hz/140 dB	0.0000	0.0001	0.0000
250 Hz/140 dB	0.0058	0.0760	0.0021
	cod adult		
25 Hz/130 dB	0.0023	0.0000	0.0000
25 Hz/140 dB	0.0138	0.3805	0.0013
60 Hz/130 dB	0.0000	0.0003	0.4839
60 Hz/140 dB	0.0000	0.0000	0.0003
90 Hz/130 dB	0.0037	0.0157	0.0470
90 Hz/140 dB	0.0000	0.0140	0.0000
125 Hz/130 dB	0.2319	0.1440	0.1012
125 Hz/140 dB	0.4288	0.0066	0.0000
250 Hz/140 dB	0.0000	0.0825	0.0000

Table A 6: Statistical results (Kruskal-Wallis-test, Mann-Whitney U-Test) for significant differences in cod numbers in tank quarter 4 between the periods before, during and after sound production. Sound levels in dB re 1µPa.

	Kruskal-Wallis test	Mann-Whitney test		
Sound situation	before sound/ during sound/ after sound	before sound/ during sound	during sound/ after sound	before sound/ after sound
cod juvenile				
25 Hz/130 dB	0.2249	0.1074	0.3552	0.0473
25 Hz/140 dB	0.0000	0.0000	0.0000	0.0872
60 Hz/130 dB	0.0000	0.0000	0.0001	0.0038
60 Hz/140 dB	0.0000	0.0000	0.0000	0.3663
90 Hz/130 dB	0.0000	0.1365	0.0000	0.0000
90 Hz/140 dB Quarter 1	0.0000	0.0000	0.0000	0.0000
90 Hz/140 dB Quarter 4	0.0000	0.0000	0.0000	0.0000
125 Hz/130 dB	0.0000	0.0000	0.0001	0.0001
125 Hz/140 dB	0.0000	0.0123	0.0000	0.0000
250 Hz/140 dB	0.0006	0.0541	0.0000	0.0235
cod adult				
25 Hz/130 dB	0.0000	0.0000	0.0000	0.2015
25 Hz/140 dB	0.0000	0.1946	0.0000	0.0000
60 Hz/130 dB	0.0000	0.0916	0.0000	0.0000
60 Hz/140 dB	0.0000	0.0000	0.0000	0.2130
90 Hz/130 dB	0.0000	0.0000	0.0000	0.0790
90 Hz/140 dB	0.0000	0.0000	0.3081	0.0000
125 Hz/130 dB	0.7589	0.4084	0.3008	0.2331
125 Hz/140 dB	0.0000	0.0000	0.0007	0.0013
250 Hz/140 dB	0.6437	0.2995	0.2887	0.1935

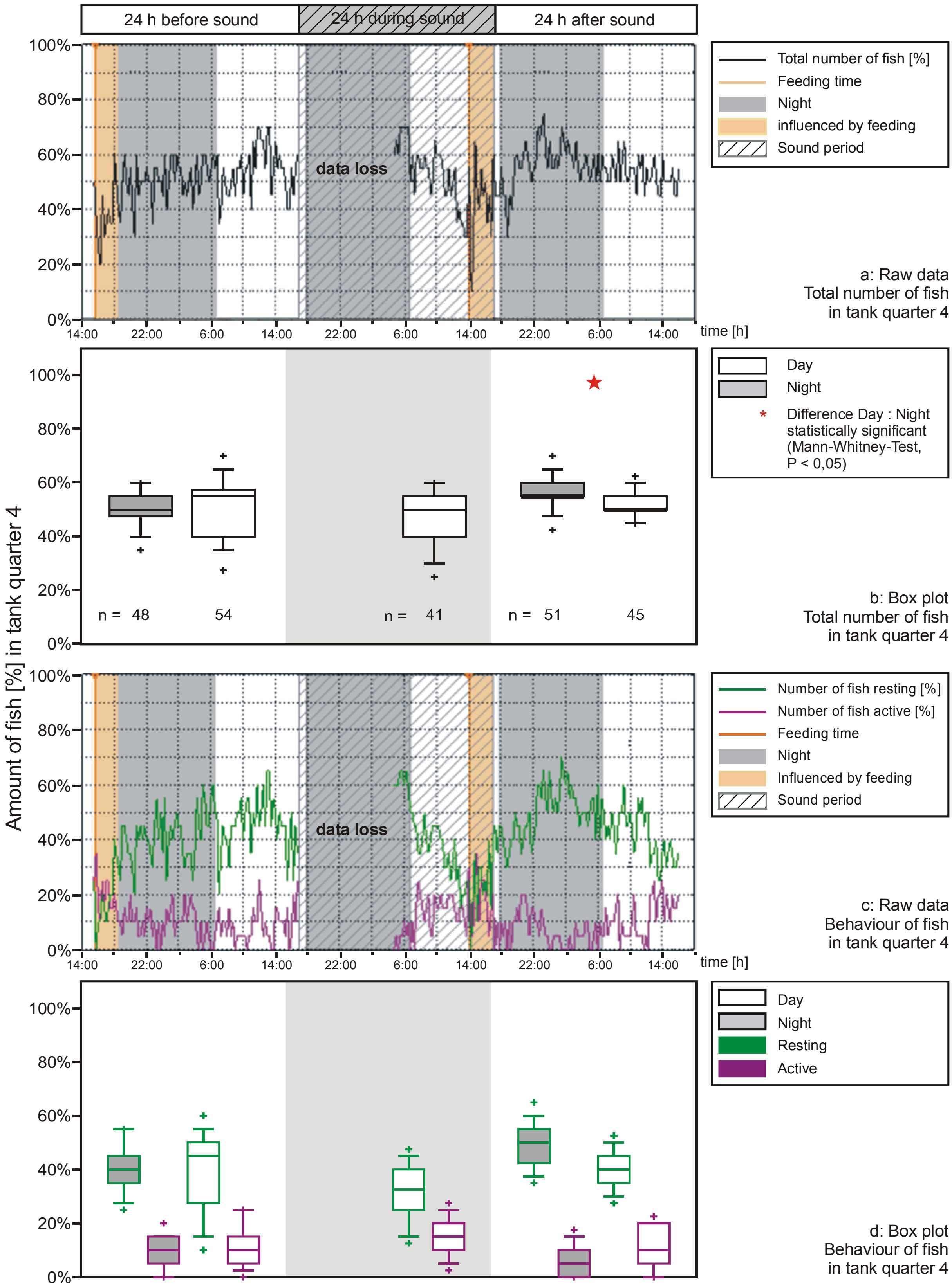

Fig. A 45: Results of sound experiment: Juvenile plaice, 25 Hz, 130 dB re 1µPa.

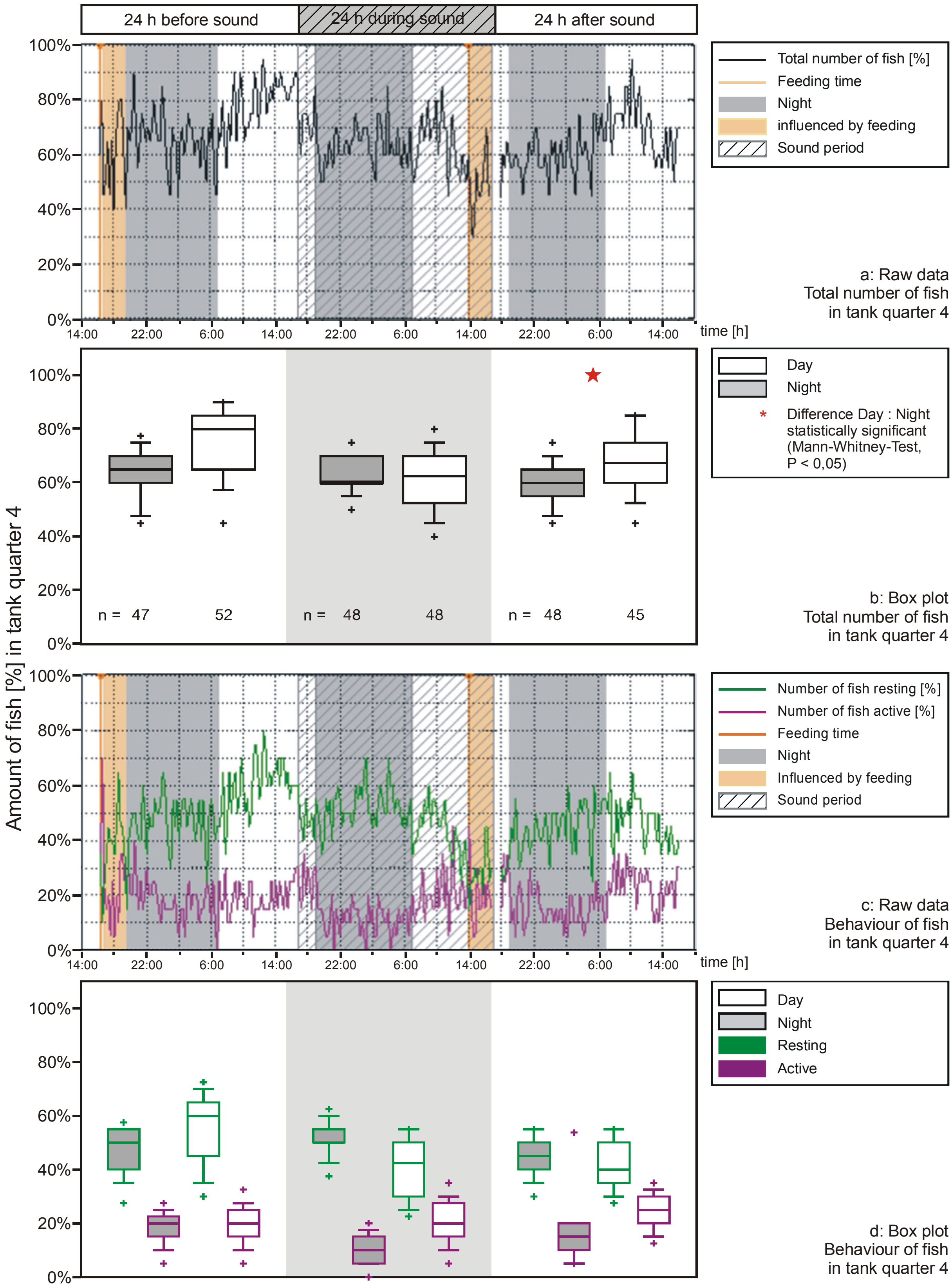

Fig. A 46: Results of sound experiment: Juvenile plaice, 60 Hz, 130 dB re 1µPa.

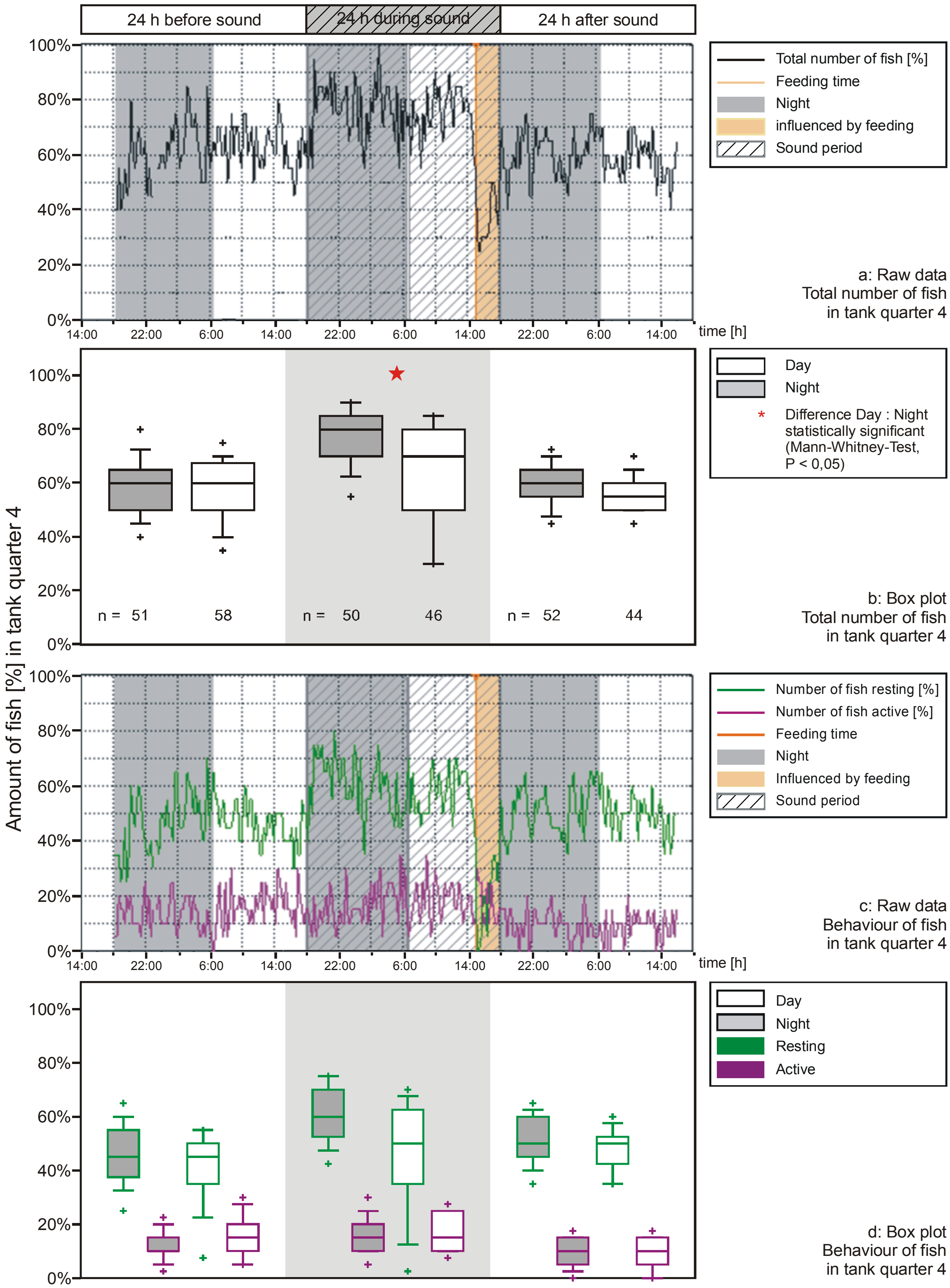

Fig. A 47: Results of sound experiment: Juvenile plaice, 60 Hz, 140 dB re 1µPa.

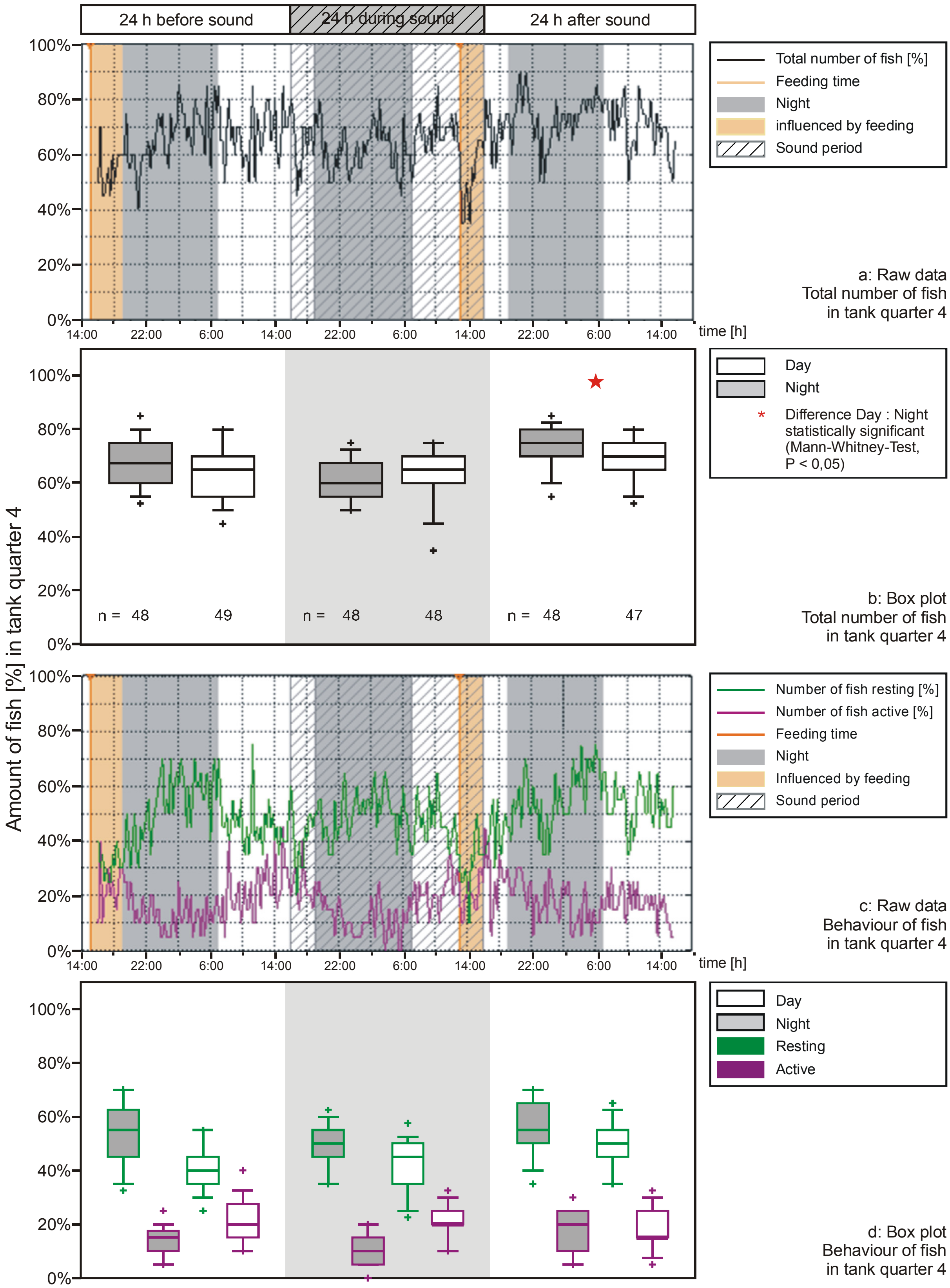

Fig. A 48: Results of sound experiment: Juvenile plaice, 90 Hz, 130 dB re 1µPa.

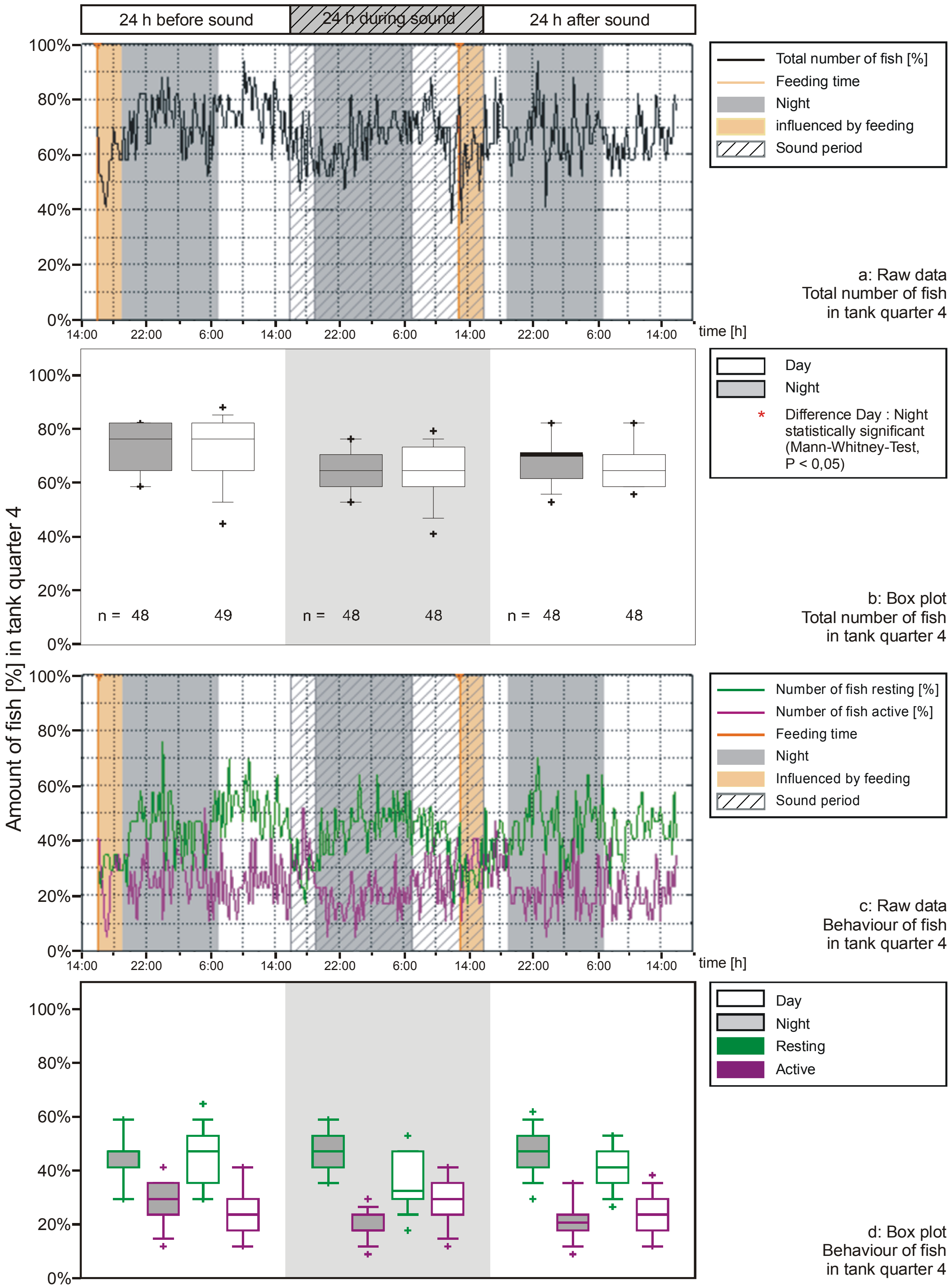

Fig. A 49: Results of sound experiment: Juvenile plaice, 90 Hz, 140 dB re 1µPa.

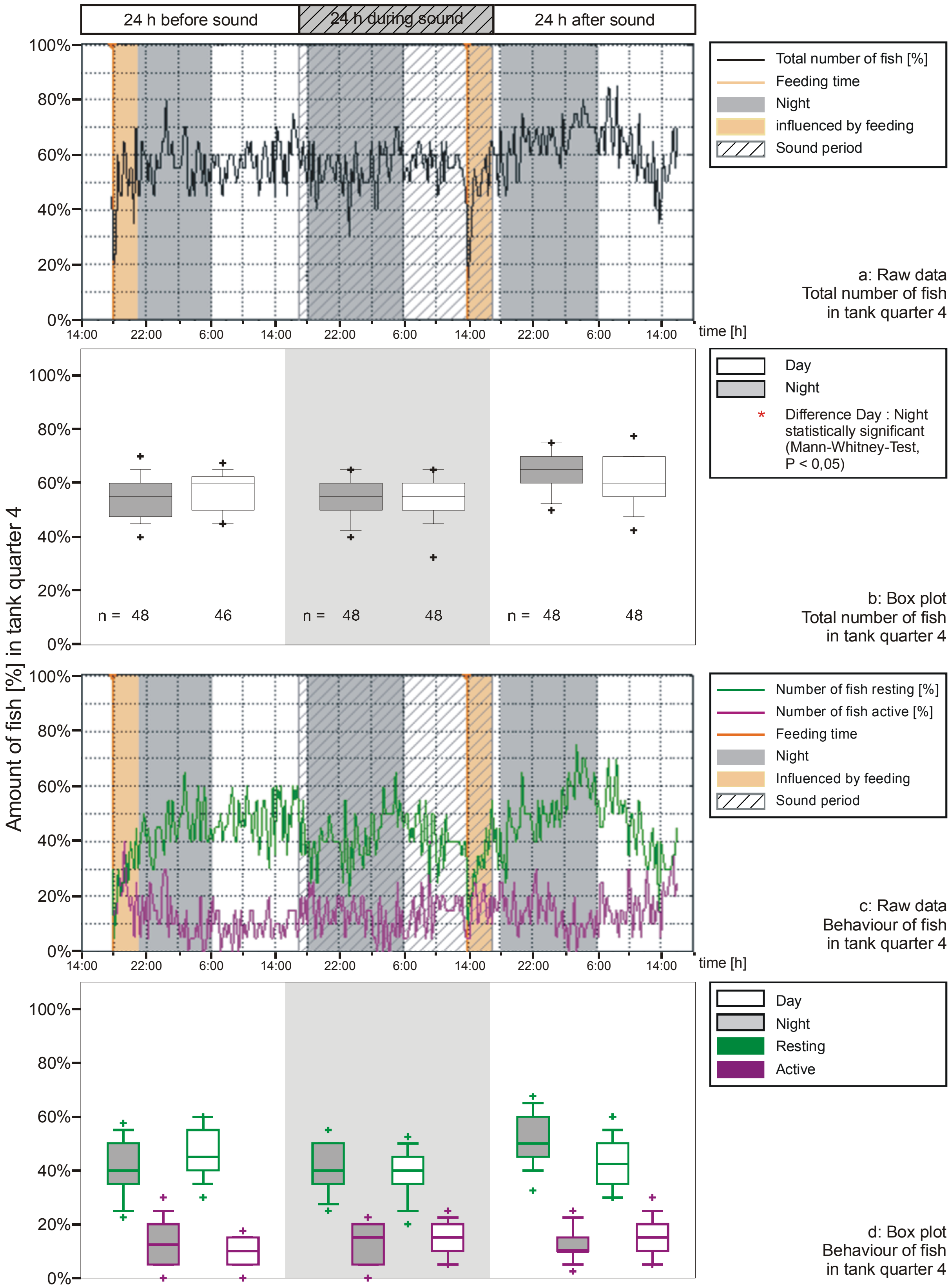

Fig. A 50: Results of sound experiment: Juvenile plaice, 125 Hz, 130 dB re 1µPa.

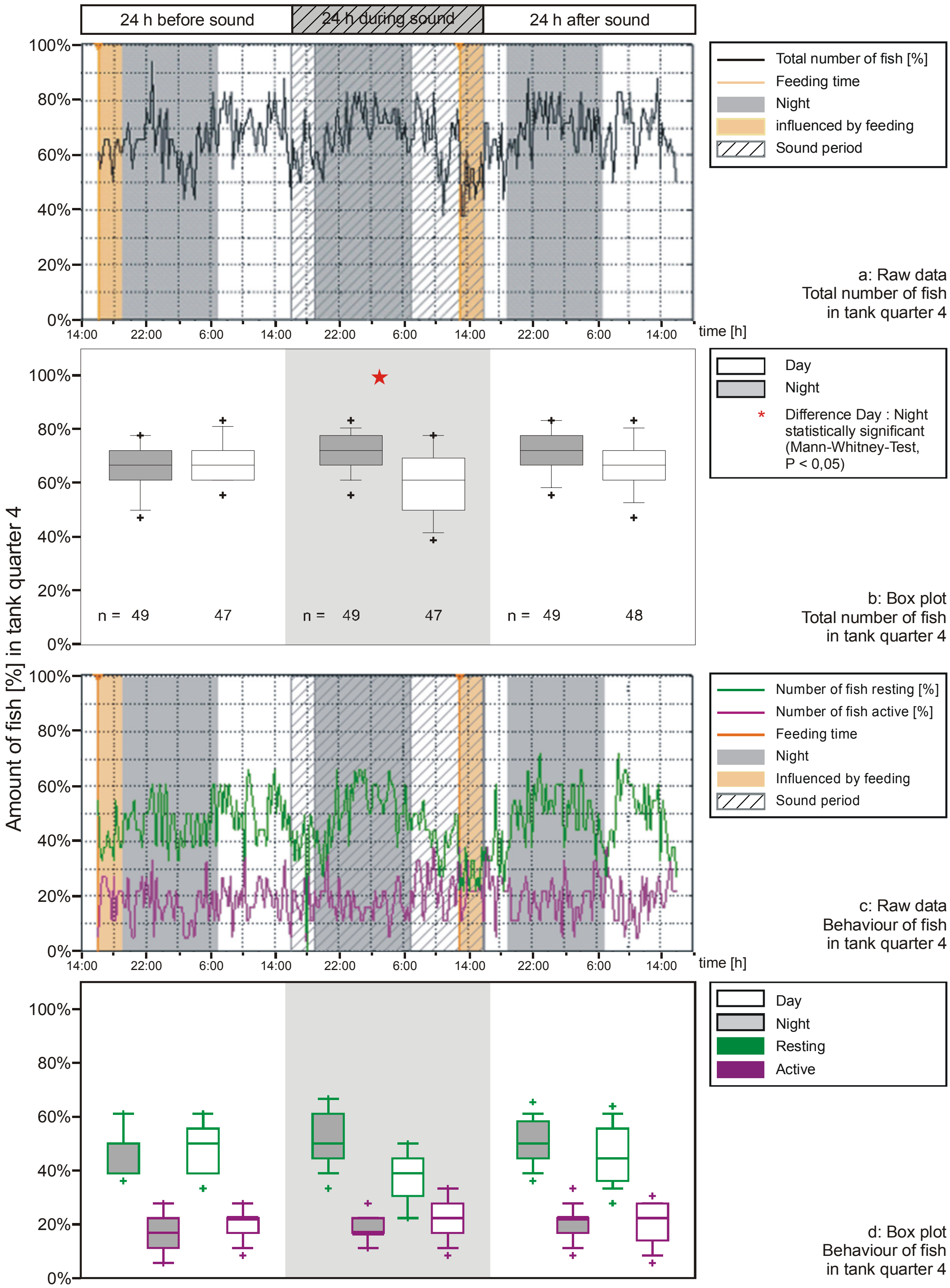

Fig. A 51: Results of sound experiment: Juvenile plaice, 125 Hz, 140 dB re 1µPa.

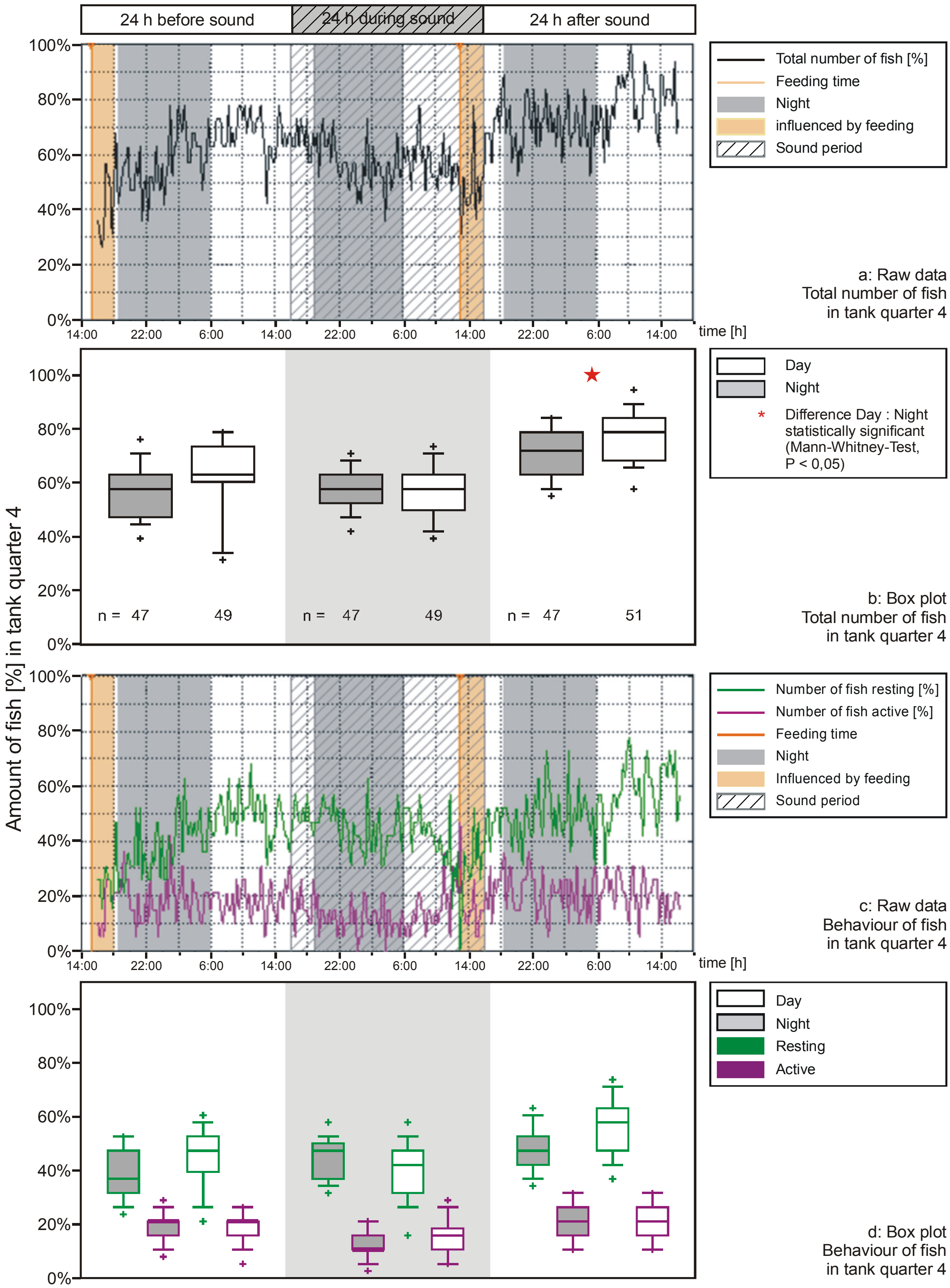

Fig. A 52: Results of sound experiment: Juvenile plaice, 250 Hz, 130 dB re 1µPa.

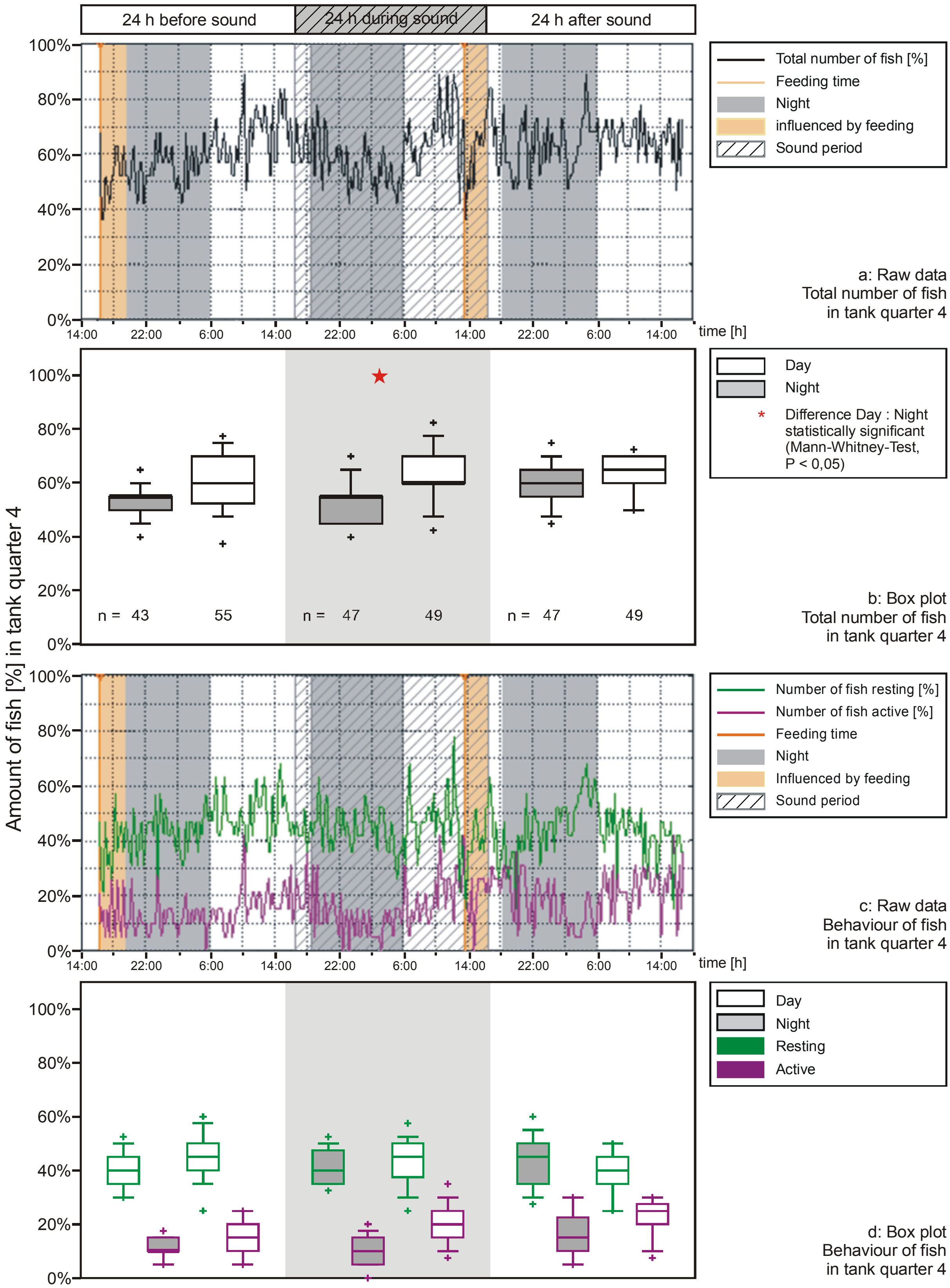

Fig. A 53: Results of sound experiment: Juvenile plaice, 250 Hz ,140 dB re 1µPa.

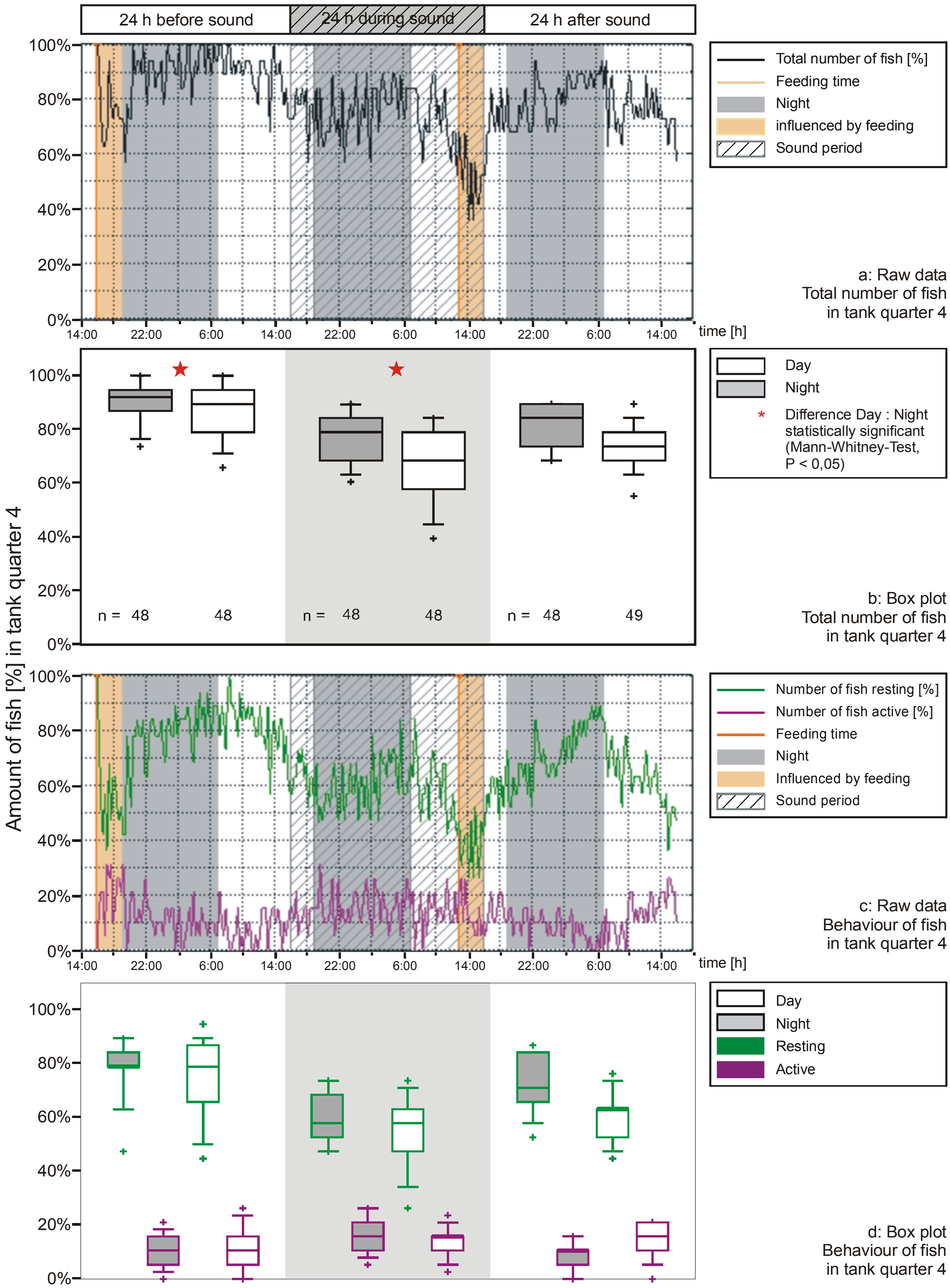

Fig. A 54: Results of sound experiment: Adult plaice, 25 Hz, 130 dB re 1µPa.

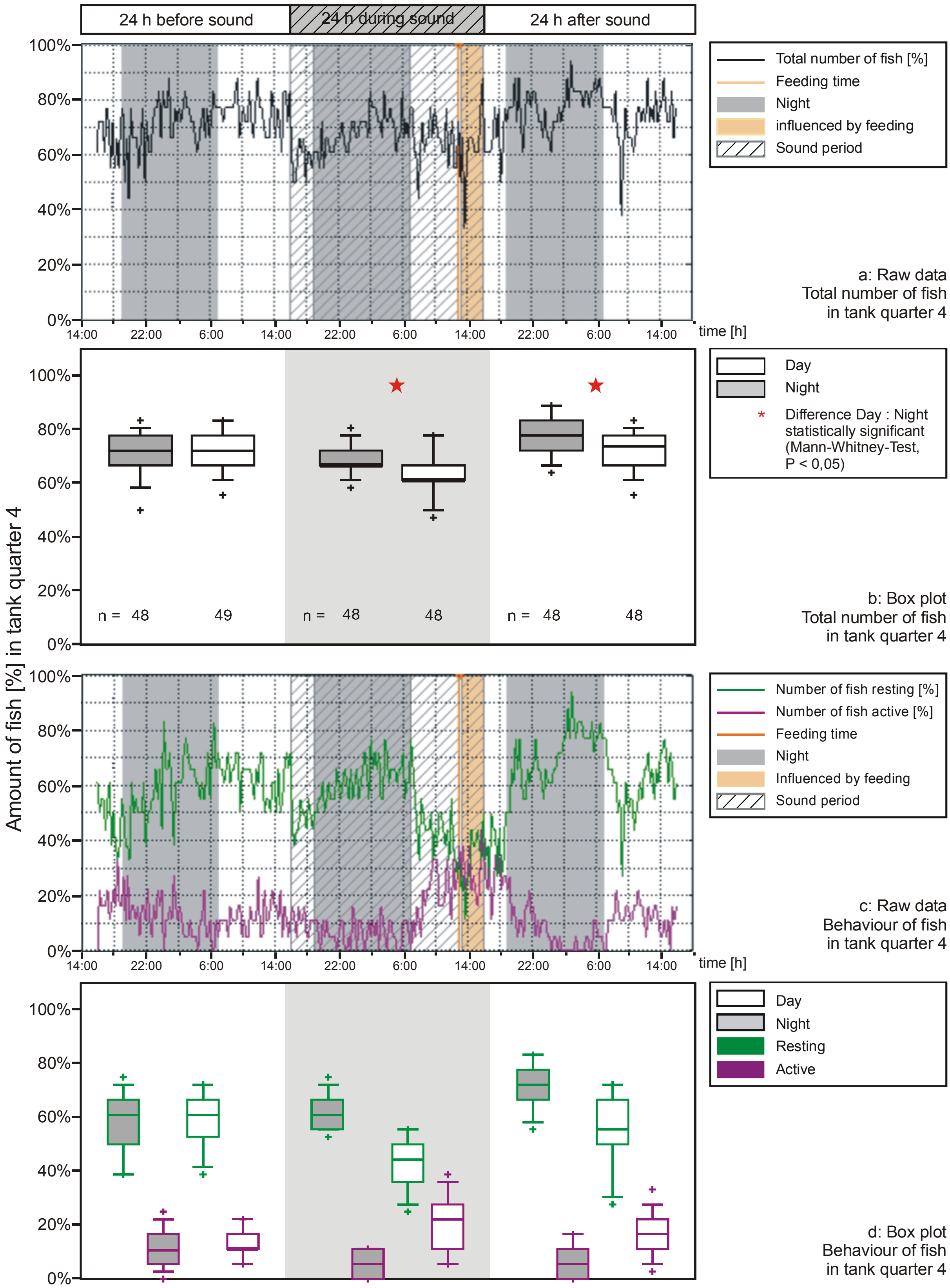

Fig. A 55: Results of sound experiment: Adult plaice, 25 Hz, 140 dB re 1µPa.

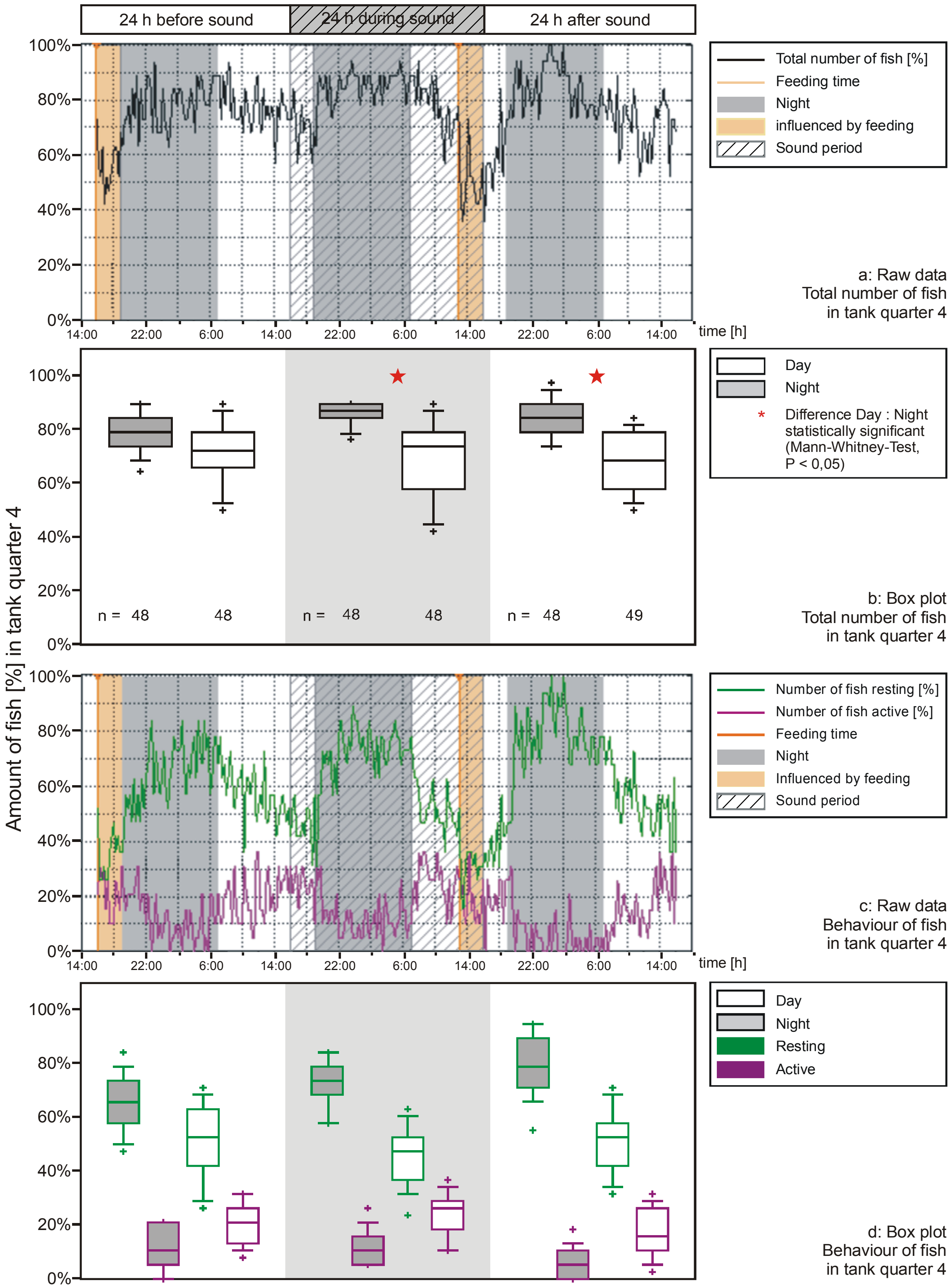

Fig. A 56: Results of sound experiment: Adult plaice, 60 Hz, 130 dB re 1µPa.

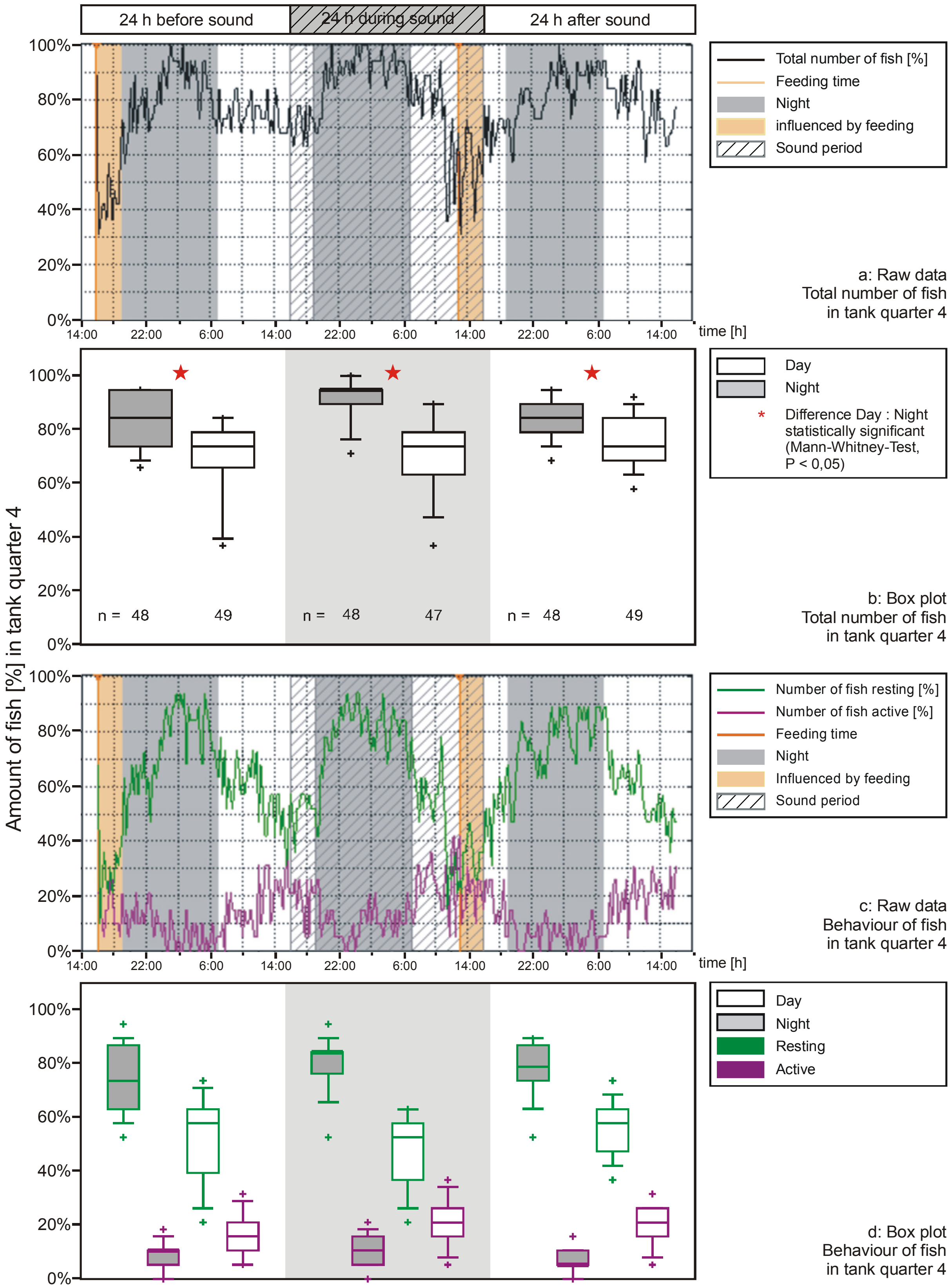

Fig. A 57: Results of sound experiment: Adult plaice, 60 Hz, 140 dB re 1µPa.

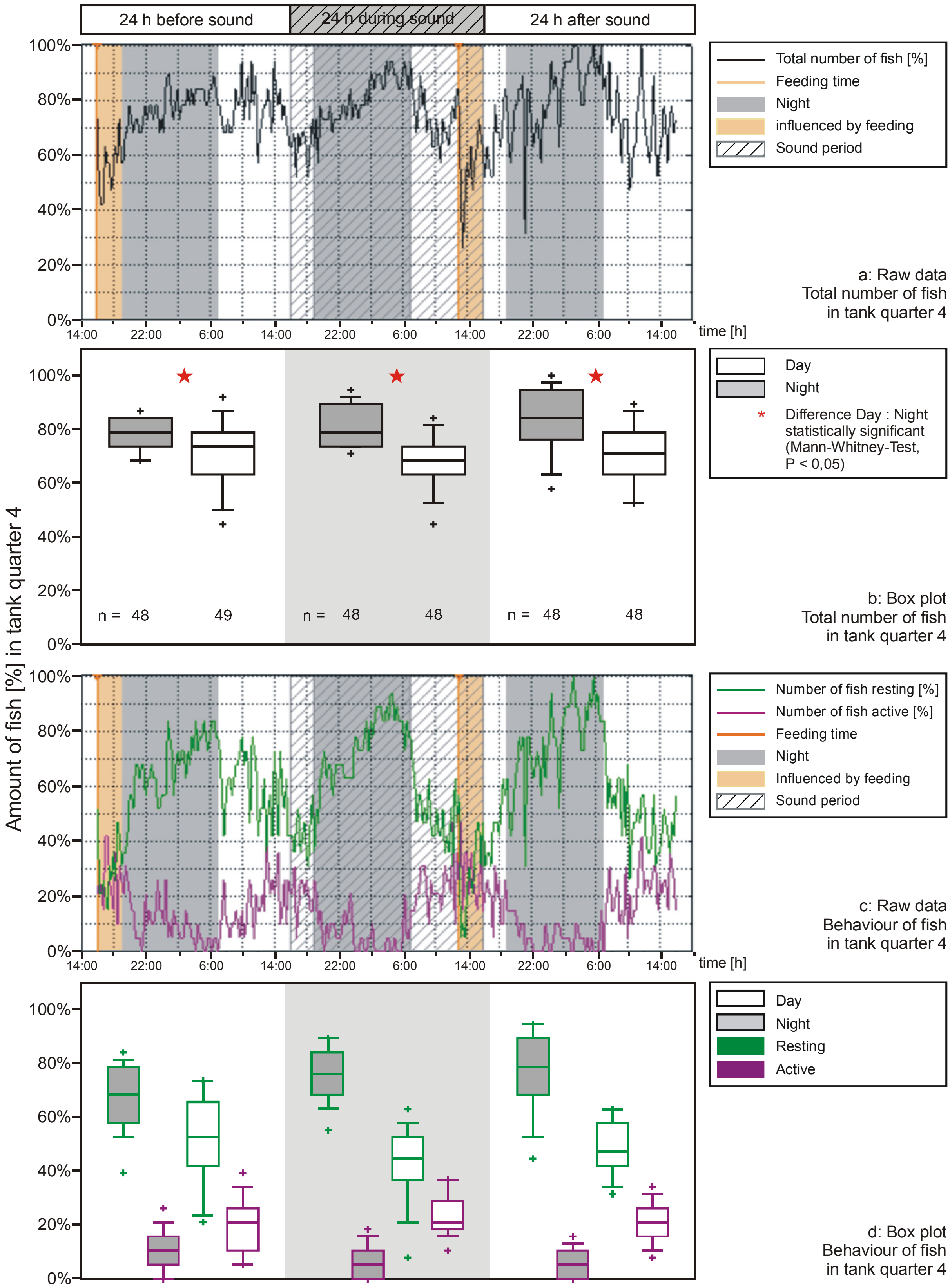

Fig. A 58: Results of sound experiment: Adult plaice, 90 Hz, 130 dB re 1µPa.

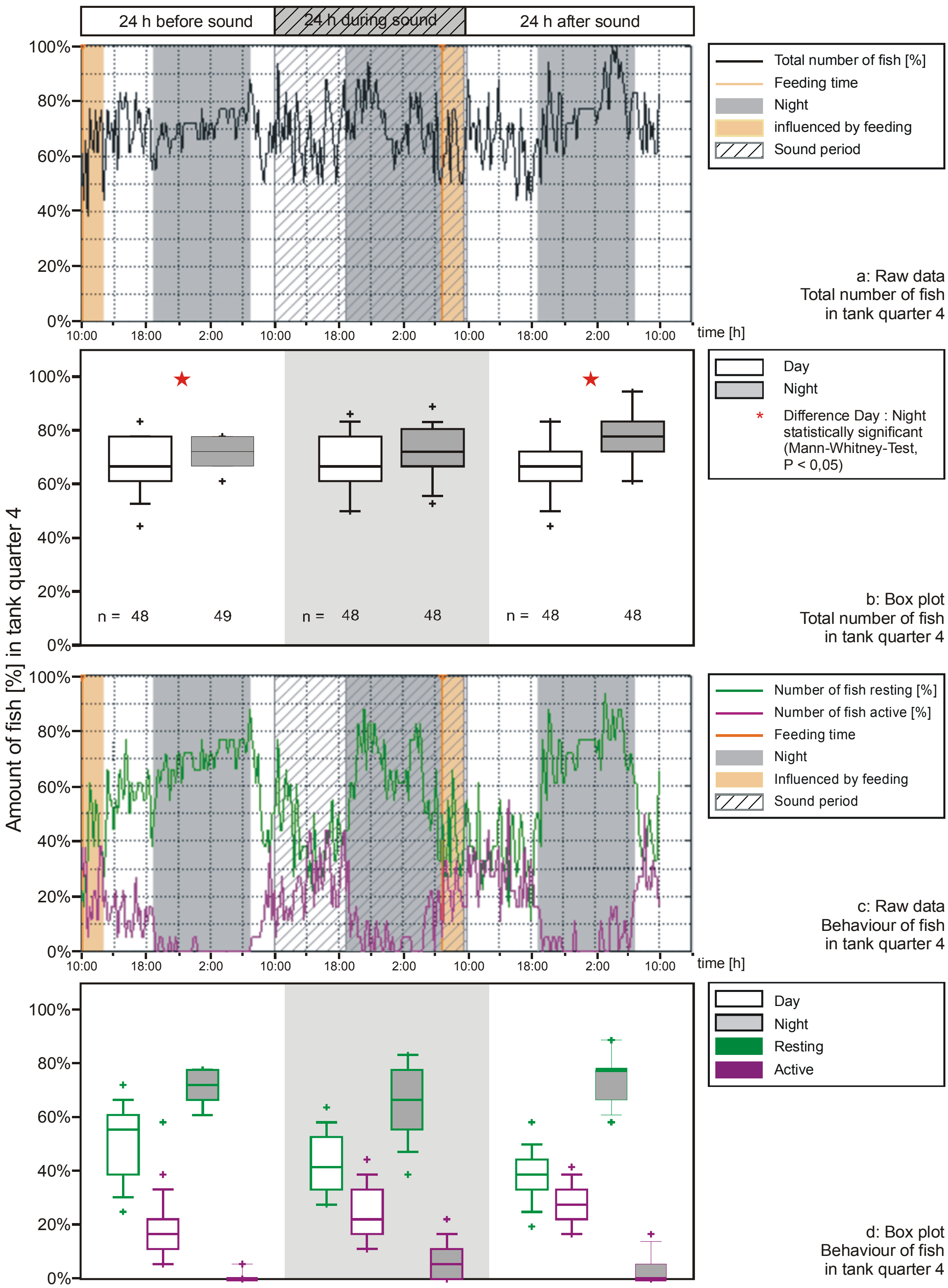

Fig. A 59: Results of sound experiment: Adult plaice, 90 Hz, 140 dB re 1µPa.

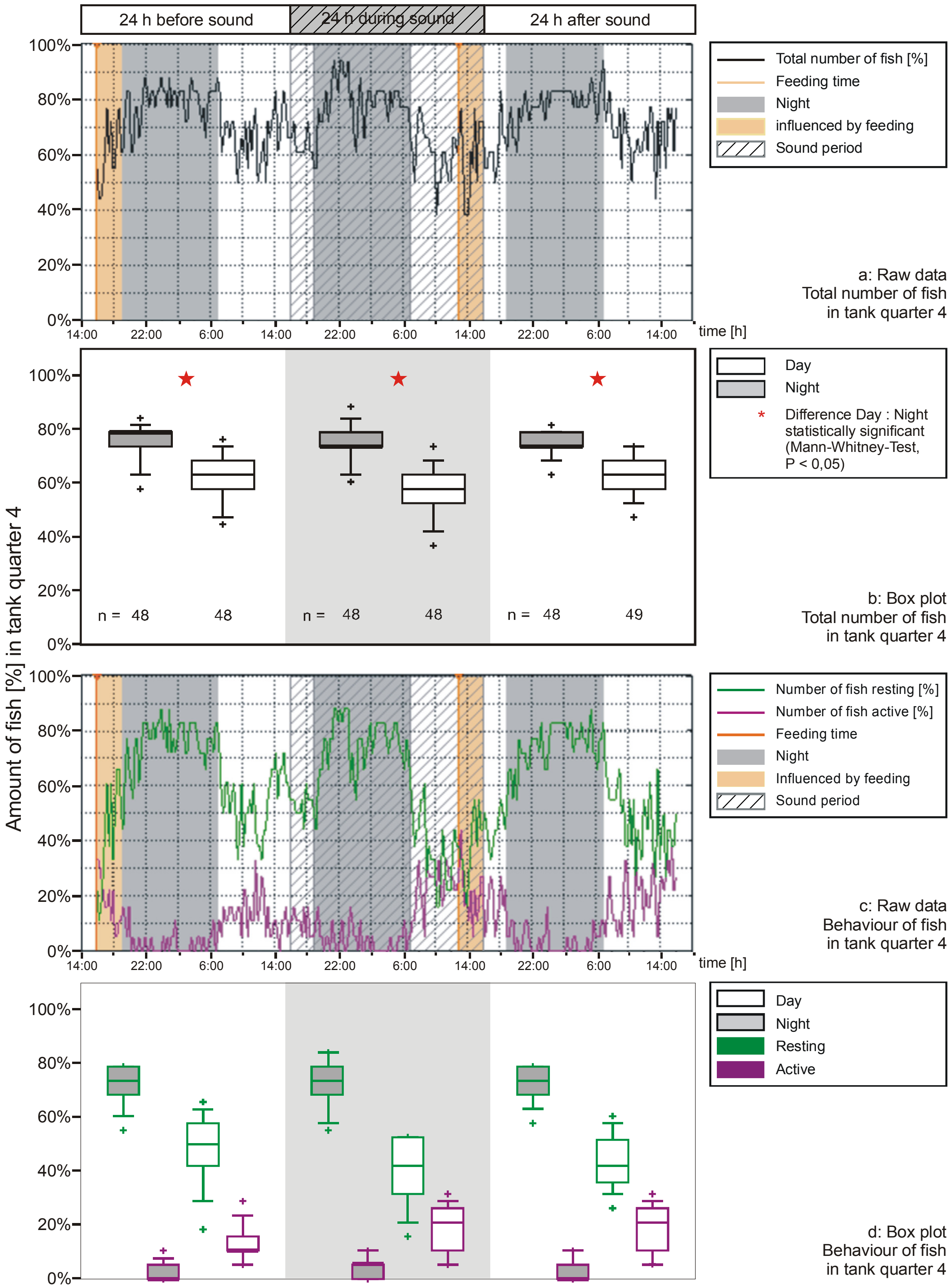

Fig. A 60: Results of sound experiment: Adult plaice, 125 Hz, 130 dB re 1µPa.

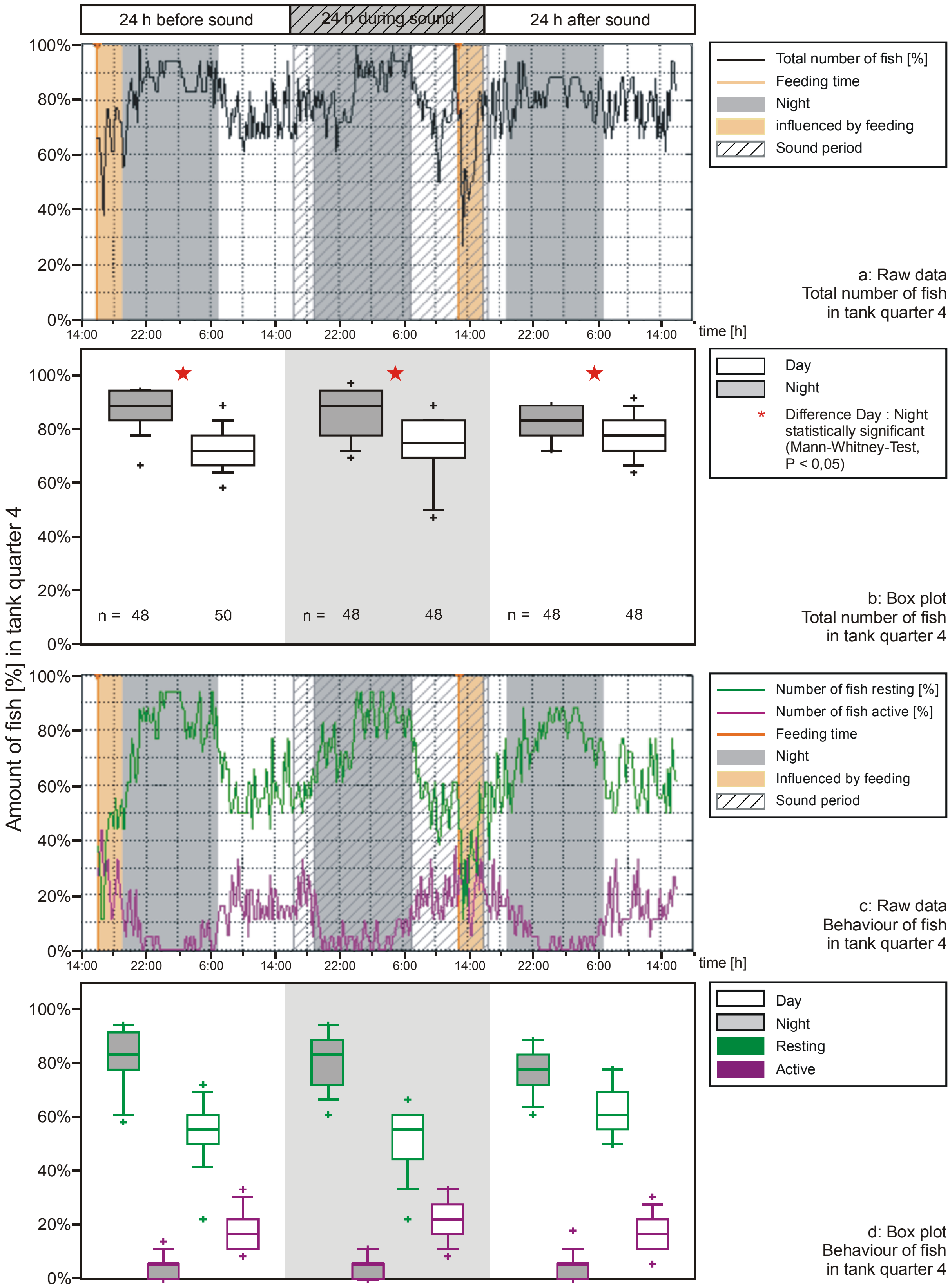

Fig. A 61: Results of sound experiment: Adult plaice, 125 Hz, 140 dB re 1µPa.

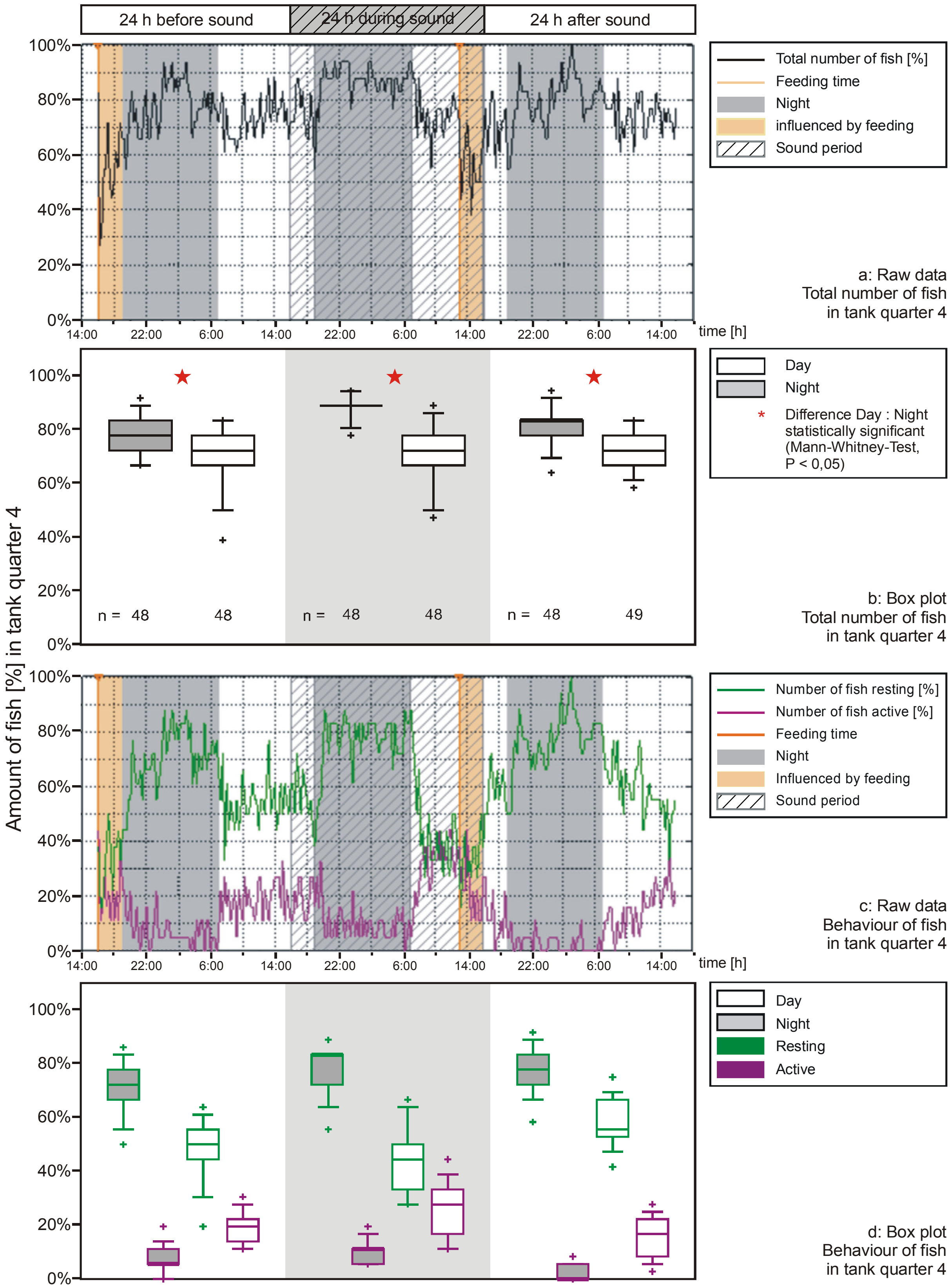

Fig. A 62: Results of sound experiment: Adult plaice, 250 Hz, 130 dB re 1µPa.

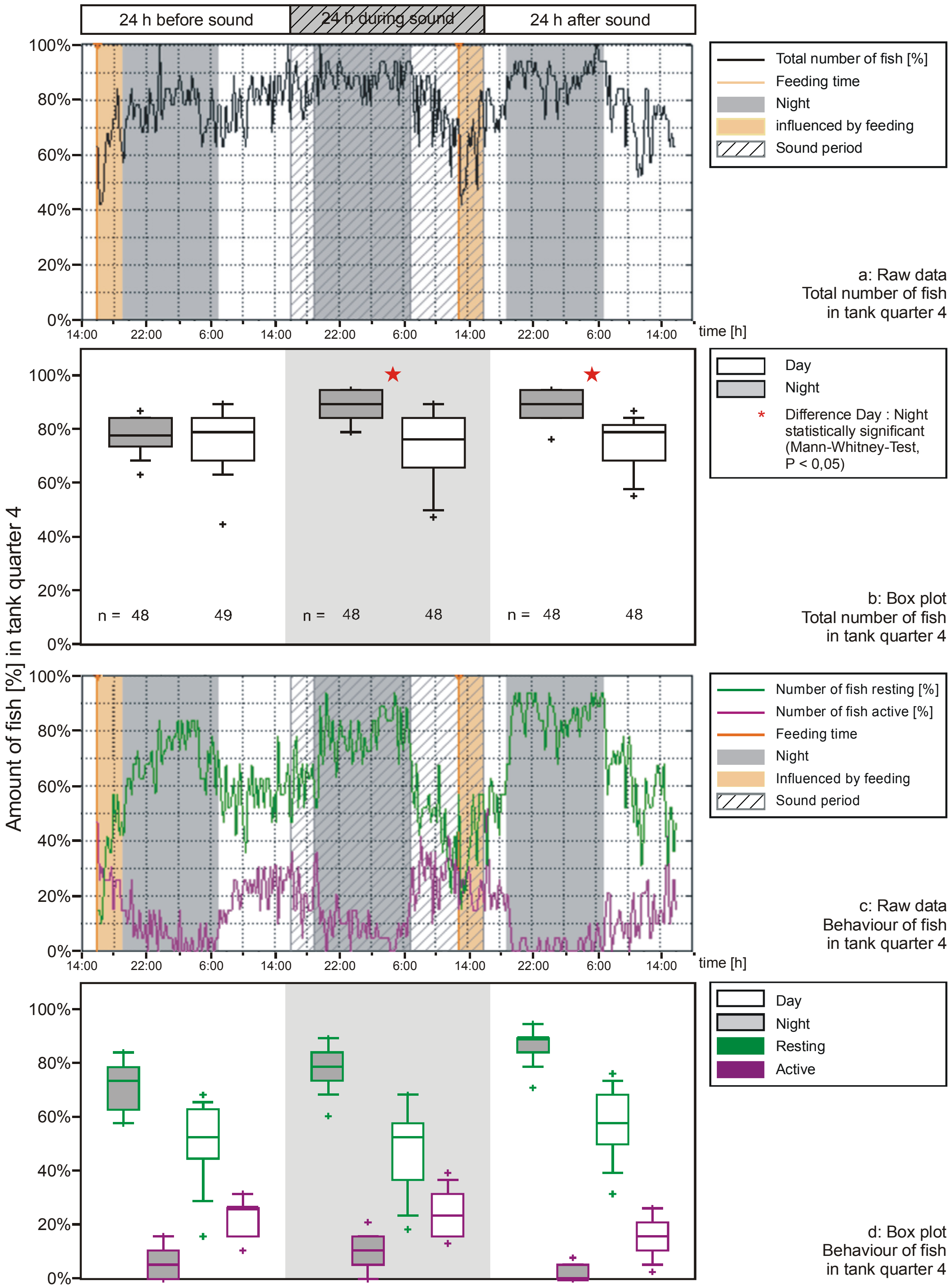

Fig. A 63: Results of sound experiment: Adult plaice, 250 Hz, 140 dB re 1µPa.

Table A 7: Statistical results (Mann-Whitney U-Test) for significant differences in plaice numbers in tank quarter 4 between day and night during the periods before, during and after sound production. Results based on one observation per hour. Sound levels in dB re 1µPa.

Sound situation	before sound	during sound	after sound
		plaice juvenile	
25 Hz/130 dB	0.2658		0.0007
60 Hz/130 dB	0.0710	0.4805	0.0232
60 Hz/140 dB	0.4807	0.0164	0.1924
90 Hz/130 dB	0.1966	0.2710	0.0266
90 Hz/140 dB	0.0859	0.1361	0.1854
125 Hz/130 dB	0.2734	0.1528	0.1632
125 Hz/140 dB	0.1471	0.0069	0.0874
250 Hz/130 dB	0.4036	0.2710	0.0499
250 Hz/140 dB	0.2008	0.0438	0.2627
		plaice adult	
25 Hz/130 dB	0.0302	0.0438	0.0592
25 Hz/140 dB	0.4708	0.0060	0.0124
60 Hz/130 dB	0.1067	0.0001	0.0015
60 Hz/140 dB	0.0159	0.0001	0.0018
90 Hz/130 dB	0.0039	0.0001	0.0001
90 Hz/140 dB	0.0102	0.1707	0.0025
125 Hz/130 dB	0.0000	0.0002	0.0002
125 Hz/140 dB	0.0001	0.0008	0.0149
250 Hz/130 dB	0.0255	0.0000	0.0025
250 Hz/140 dB	0.2321	0.0025	0.0000

Table A 8: Statistical results (Kruskal-Wallis, Mann-Whitney U-Test) for significant differences in plaice numbers in tank quarter 4 between the periods before, during and after sound production. Results based on one observation per hour. Sound levels in dB re 1µPa.

	Kruskal-Wallis test	Mann-Whitney test		
Sound situation	before sound/ during sound/ after sound	before sound/ during sound	during sound/ after sound	before sound/ after sound
plaice juvenile				
25 Hz/130 dB	0.0861			
60 Hz/130 dB	0.0367	0.4887	0.0352	0.0150
60 Hz/140 dB	0.0004	0.0004	0.0012	0.2396
90 Hz/130 dB	0.2583			
90 Hz/140 dB	0.0009	0.0023	0.1139	0.0029
125 Hz/130 dB	0.0014	0.1953	0.0020	0.0001
125 Hz/140 dB	0.8013			
250 Hz/130 dB	0.0002	0.3670	0.0001	0.0006
250 Hz/140 dB	0.0476	0.4812	0.0055	0.0185
plaice adult				
25 Hz/130 dB	0.0000	0.0000	0.0484	0.0001
25 Hz/140 dB	0.0567			
60 Hz/130 dB	0.0344	0.1379	0.2743	0.4880
60 Hz/140 dB	0.0614			
90 Hz/130 dB	0.9999			
90 Hz/140 dB	0.5680			
125 Hz/130 dB	0.6462			
125 Hz/140 dB	0.9334			
250 Hz/130 dB	0.1150			
250 Hz/140 dB	0.0949			

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MÜLLER-BLENKLE, C., REID, D., JONES, E., LÜDEMANN, K., KAFEMANN, R., ELEPFANDT, A.: Influence of low frequency sound resembling offshore wind turbine noise on fish. Conference Proceedings 19th International Congress on Acoustics (in print).

Eidesstattliche Erklärung

Ich versichere hiermit, die vorliegende Arbeit selbständig verfasst und keine anderen Hilfsmittel und Quellen als die angegebenen verwendet zu haben.

Berlin, 25. Juli 2007